T 28

United States
Environmental Protection
Agency

Office of Pesticides
and Toxic Substances
Washington, DC 20460

EPA-560/11-80-027
October 1980

Toxic Substances

EPA

Support Document Test Data Development Standards

Physical/Chemical and Persistence Characteristics:

Density/Relative Density
Melting Temperatures
Vapor Pressure
Octanol/Water Partition Coefficient
Soil Thin Layer Chromatography

Proposed Rule, Section 4

Toxic Substances Control Act

NEWARK PUBLIC LIBRARY

NOV 10 1980

S. GOVT. PUBS.
DEPOSITORY 376

EP 5.2:

EPA 560/11-80-027

SUPPORT DOCUMENT
TEST DATA DEVELOPMENT STANDARDS
PHYSICAL/CHEMICAL & PERSISTENCE CHARACTERISTICS:

DENSITY/RELATIVE DENSITY
MELTING TEMPERATURES
VAPOR PRESSURE
OCTANOL/WATER PARTITION COEFFICIENT
SOIL THIN LAYER CHROMATOGRAPHY

PROPOSED RULE, SECTION 4
TOXIC SUBSTANCES CONTROL ACT

Exposure Evaluation Division
Office of Toxic Substances
OCTOBER 1980

U. S. Environmental Protection Agency
Office of Pesticides & Toxic Substances
Washington, D.C. 20460

TABLE OF CONTENTS

[Numbers Refer to Those at Top of Page]

Density/Relative Density..... 1

Melting Temperatures..... 27

Vapor Pressure..... 51

Octanol/Water Partition
Coefficient..... 79

Soil Thin Layer
Chromatography.....121

DENSITY/RELATIVE DENSITY

STANDARD FOR TEST DATA DEVELOPMENT
PROPOSED RULE, SECTION 4, TSCA

Refers to

Part 772 -- Standards for Development of Test Data
Subpart I. Physical, Chemical and Environmental
Persistence Characteristics
Section 772.122-1 Density/Relative Density

October 1980

CONTENTS

Page

1. NEED FOR THE TEST..... 1

2. SCIENTIFIC ASPECTS..... 4

2.1. Selection of Test Conditions..... 4

2.1.1. Equipment..... 4

2.1.2. Materials..... 5

2.2. Selection of Test Procedures..... 5

2.2.1. Temperature of Test..... 5

2.2.2. Selection of Measurement Technique... 6

2.2.3. Calculation of Ideal Gas Density.....10

2.3. Test Data Required.....10

3. ECONOMIC ASPECTS.....12

4. COMPATABILITY WITH OTHER TEST STANDARDS.....13

4.1. TSCA/FIFRA Compatibility.....14

4.2. OECD Compatibility.....14

4.3. IRLG Compatibility.....14

5. RESPONSES TO COMMENTS RECEIVED ON PROPOSED GUIDANCE FOR PREMANUFACTURE TESTING.....15

5.1. General.....15

5.2. Responses to Specific Comments.....15

6. REFERENCES.....19

1. NEED FOR THE TEST

The purpose of this test is to develop data on density and relative density (specific gravity) of chemical substances and mixtures.

EPA will use the data obtained from testing of density and relative density to evaluate the potential for human and environmental exposure to chemicals. Data also will be used as a screening test in order to determine what additional testing will be required for physical and chemical characteristics and for effects on the various portions of the living and nonliving environment.

Density is an important factor affecting the path that a chemical will take in moving through the environment and affecting where the material may eventually accumulate. As chemical substances enter the atmosphere or water, as a result of normal practices of manufacture, transportation, use, ultimate disposal, or accidental spill, they will tend to rise, sink, or disperse as a function of their density. In water, for example, low density substances (liquid or solid) will float to the surface, dense substances will tend to sink to the bottom, and materials with approximately the same density as the water will tend to become dispersed. EPA, therefore will use information on density to determine if principal exposure may be to bottom-dwelling organisms (e.g., shellfish), to plants or animals (e.g., waterfowl) that live at the surface, to swimmers who use the shallow beach areas, or to swimming fish which contact the

dispersed chemicals. The information will be used in selecting which types of plants or animals to test for toxic effects, and it will be used in the ultimate evaluation of risk to the human and nonhuman environment.

Similar considerations need to be made for materials that enter the atmosphere through any of a number of ways, intentional or accidental. Dense gases will tend to accumulate near the surface of the earth. An example is chlorine, which occasionally is spilled as a result of train, barge, or truck wrecks or the rupture of storage vessels. Because of its density, the gas remains near the surface and causes a great risk to nearby human and animal populations. Light gases, on the other hand, tend to rise and diffuse in the atmosphere where they may be subject to photochemical degradation. However, if they are not degraded they may be transported great distances and ultimately be redeposited through various processes of atmospheric deposition.

One way that EPA will use data from density testing is as part of the input to several mathematical models for assessing transport and fate of chemicals in the environment. These models, which consider movement in the atmosphere, surface waters, and ground water, all require information on density to evaluate the movement of materials, their ultimate deposition, and the biological and chemical processes of transformation that

will affect them. Results from the modeling work are then combined with the results of toxicity and other hazard evaluations in order to assess the total human and environmental risk from the chemicals.

2. SCIENTIFIC ASPECTS

2.1. Selection of Test Conditions

2.1.1. Equipment

The use of calibrated ASTM-specification thermometers is required. The particular type of thermometer is usually specified in the standard technique identified in Table 1 of the Test Standard.

Widespread laboratory experience has found that manufactured thermometers are sometimes inaccurate by several degrees. Such inaccuracy may result in incorrect density values that would adversely affect EPA's ability to use the data in models. A minimal, satisfactory, easily attainable traceability to NBS is a comparison by the manufacturer or the analyst with a thermometer that has been calibrated by NBS. Compliance with this condition of the test is relatively easy, but it is an important factor that often is overlooked.

The other apparatus required is fully described in the methods and procedures referenced in Table 1 of the Standard. EPA has reviewed and adopted these procedures with their appropriate equipment requirements.

2.1.2. Materials

The reference materials, water, mercury, cyclohexane, and toluene have been recommended by the IUPAC Commission on Physicochemical Measurements and Standards (1975, 1976), and recommended density values have been given. These, or other appropriately selected substances, are to be used in this procedure for calibrating the apparatus as described in several of the techniques.

The suggested immersion liquids are water and a series of organic compounds useful in the event that the substance is soluble in water. The liquids listed are representative and have been recommended in one or more of the techniques requiring immersion of the specimen. The immersion liquids include several of high density, to be used in the sink-float technique. The comparison gases are for use with the gas-comparator pycnometer technique.

2.2. Selection of Test Procedures

2.2.1. Temperature of test

The IUPAC has for many years recommended (IUPAC 1972) the reporting of physical-chemical-properties measurements at the convenient temperature of 25°C, and this is the preferred temperature. Testing at this temperature will provide the reliable data that EPA requires for use in its modeling techniques for evaluating environmental fate and exposure. A

temperature of 25°C is slightly above customary laboratory temperature, which makes for convenient adjustment and maintenance of a constant temperature bath at 25°C for measurements.

However, engineering groups, such as the American Petroleum Institute and the American Gas Association have established a much lower reference temperature. Their most common reference temperature for many years had been 60°F (15.55°C). Within the last few years, with the shift to SI units, and to harmonize their standards with the European standards (through ISO) they have agreed to shift to 15°C as the reference temperature. This temperature is representative of many environmental temperatures and EPA will be able to adjust its evaluating techniques to use density data determined at 15°C.

The CIPAC group has adopted (Raw 1970) 20°C as the standard reporting temperature for the properties of pesticides. A substantial body of data on pesticides has been determined at this temperature and the results are useable by EPA. Pesticides are commonly used at temperatures which are somewhat warmer than annual temperature because they are generally used during the growing season or indoors.

2.2.2. Selection of a Measurement Technique

The procedure for testing chemicals under the Toxic Substances Control Act (TSCA) must be broad enough to cover the entire range of chemicals subject to the law. Measurement of

density, in itself, is not a complicated problem for most chemicals, however, the techniques for handling different kinds of chemicals are quite different because of different physical characteristics.

Many simple procedures have been developed for making measurements on common classes of chemicals. These are based on well known physical principles, such as Archimedes' principle (see, for example Bauer and Lewin [1971], p.91) or on the definition of density itself. The different techniques have specific limitations because of the shapes of the measuring devices, the incompatibility of certain chemicals with immersion liquids, the viscosity and the volatility of the test substance, or the size of the particles that may have to be tested.

EPA has reviewed a broad range of methods from the scientific literature and has adopted the large number of standardized protocols listed in Table 1 of the Standard. Each method has been evaluated for suitability for use with different types of substances as described by characteristics given in Table 2 of the Standard. Section (e)(1)(ii) of the Standard specifies which types of techniques EPA has judged suitable for use with the different types of substances. The person doing the testing then is free to choose from among the appropriate standardized protocols listed in Table 1.

Some contingencies may not be covered by the identified techniques in this procedure. In these cases the method to be used will be specified in the test rule.

An important feature of Table 1 is its application to the choice of a technique that is suitable to the physical characteristics of the material to be tested. For assistance in this process, Table 2 of the Test Standard has been developed to give the important characteristics that must be taken into account in making the selection of a technique. If further guidance is necessary, EPA recommends the book by Bauer and Lewin (1971), Table 2.7, which outlines recommended techniques for various special problems.

For gaseous substances the principal decision is whether to calculate or measure the density. In consideration of the relatively small differences between results produced by the two techniques, the Agency will accept a calculation based on the ideal gas law as adequate. This method is suitable for assessment purposes, as indicated by a comparison of real gas and ideal gas densities of several typical industrial gases (allene, ammonia, 1,3-butadiene, chlorine, methylamine, helium, fluoromethane, and nitrous oxide), at atmospheric pressure and ambient temperatures. The real gas density is higher than the ideal gas density in all cases but one. However, the average relative error is only 3.0 percent, with a standard deviation in the relative error of 4.7 percent. However, because easily used devices are available commercially for measuring gas density, [ANSI/ASTM D 1070] the option is given of measuring rather than calculating the density.

For liquids, the volatility and viscosity are factors in the selection of a method. The sensitivity of the techniques to these factors is not very great, however, so for certain ranges of volatilities and viscosities, one technique or another may work equally well. For volatilities, this range is defined by 15 kPa $< P^{\circ} <$ 80 kPa, where 1 kPa = 0.00987 atm. and P° is the vapor pressure of the pure liquid at 25°C. Similarly there is a range of viscosities in which one technique or another may equally well be used. This range is defined by 15 cST $< \tau$ (25°C) $<$ 400 St where τ is the viscosity. The viscosity primarily affects the ease with which the liquid flows through capillary tubes used in some techniques. Table 2 of the Standards accounts for the range of overlap.

For solids, the size of particles, the interfacial tension with water, and the solubility in water affect the choice of method. The selection is not strongly dependent on the characteristic, however, so the Standard leaves room for the judgment of the analyst.

Solubility of the solid in water in amounts greater than 1 mass percent precludes use of water as an immersion liquid, and some other liquid must be chosen. Also, immersion methods in which the change in weight of an object is used as a measure of its density, require pieces large enough to be handled individually. Wettability is another important parameter, particularly for powders, if the density is to be determined by displacement of water.

Table 2 lists the characteristics of interest in a systematic way for convenience, but it gives limits only in some cases for which quantitative criteria seem to be useful. Within the scope of this Table and Table 1, the sponsor is expected to determine which technique to use and to apply it appropriately in making the test.

The accuracies obtainable by the standard techniques are stated in the published descriptions of the techniques. In every case they meet or exceed the requirements of the Agency. Techniques for exposure assessment used by EPA generally accept an accuracy within 5 percent as adequate, except in the region of relative density from 0.95 to 1.05. Here, in order to determine how a chemical will act in water, the relative density needs to be known to 0.01.

2.2.3. Calibration

Calibration of laboratory equipment has long been a standard requirement for testing or research. The step, while not burdensome, assures that the data will not be affected by equipment that is damaged or has drifted out of calibration.

2.2.4. Calculations of Ideal Gas Density

The density, ρ, is calculated from the formula

$$\rho = pM/RT$$

in which p is the pressure, M the molar mass, R the thermodynamic gas constant, and T the thermodynamic temperature. Using T = 298.15 K (25°C) and R = 8.3143 J $mol^{-1}K^{-1}$, and with the mass M expressed in grams and the pressure in kilopascals, the gas density can be calculated in kilograms per cubic meter.

The relative density of an ideal gas with respect to air is calculated from the ratio of the molecular weight of the gas to the average molecular weight of air. For the average molecular weight of air, we have taken 28.964 from Weast (1978-1979).

2.3. Data Required

The sponsor is allowed the option of reporting either the density or the relative density because the calculation from one to the other is simple and unambiguous. The sponsor is also asked for a statement of the technique that was chosen and for calibration information. Because, in some instances, the density of the material tested may not indicate the range of acceptable densities in a material of variable composition, a statement of the acceptable range of densities is requested.

3. ECONOMIC ASPECTS

A survey of commercial laboratories to estimate costs for performing the tests outlined in the Standard found a range of $15 to $75, with four of the five estimates having a top range of $30, and with a "best estimate" of $28 based on the survey. A cost estimate also was made by separating the protocol into components and estimating the cost of each component, including direct labor cost, overhead cost, other direct cost, general and administrative costs, and profit or fee. The protocol best estimate of cost was $72, with an estimated range of $36 to $109 based on ± 50 percent of the best estimate. An analysis of the discrepancy between the protocol estimate and the survey pointed out that the nature of the chemical to be analyzed has an impact on the cost of analysis. The physical state of the compound (e.g., liquids versus chunk or particulate solids) determines the method to be used. Different methods will cause variations in cost. The number of samples for analysis may also affect the cost per sample. Discounts are offered by some laboratories based on the number of samples to be analyzed.

The above cost estimates were made assuming that all the requirements of Good Laboratory Practice Standards, as specified in section (c) of the Density Standard, are being satisfied. Details of the cost estimates are contained in a report by Enviro Control, Inc. (1980).

4. COMPATABILITY WITH OTHER TEST STANDARDS

4.1. TSCA/FIFRA Compatibility

Methods for determining density given in FIFRA guidelines (USEPA 1978) refer to techniques as follows: CIPAC MT3; ASTM D 941-55; ASTM D 1480-62; ASTM D 1481-62; and ASTM D 1217-54. No guideline is given as to which technique should be used. All of the above techniques are offered in the present test protocol, as well as some additional techniques which may be needed because of the greater variety of chemicals to which TSCA applies. In addition instructions are given in the TSCA Standard for the selection of an appropriate technique.

The FIFRA guidelines (USEPA 1978) specify the reporting only of density for solids and only of relative density (specific gravity)* for liquids. We offer a broader option of either density or relative density for both solids and liquids. Either type of information is equally suitable for use in the type of exposure analysis used for TSCA chemicals.

The FIFRA guidelines (USEPA 1978) specify a reporting temperature of 20°C. The TSCA protocol includes that temperature

---

*The IUPAC (McGlashan et al. 1979) has recommended the term relative density in preference to specific gravity; preferring to reserve the word "specific" to refer to a quantity per unit mass, as in specific volume. The term "specific gravity," with the same meaning as relative density is therefore obsolescent. This practice has been adopted also by ASTM (1978) for their standards.

among three options, all of which are used to some extent in nationally and internationally standardized tests. The 20°C reference temperature is adopted by CIPAC (Raw 1970), and this provides the justification for its use. However, as explained in Section III, there is substantial justification for allowing other reporting temperatures. In summary, the TSCA protocol is compatible with the FIFRA guidelines (and with proposed revisions) but is somewhat broader in scope and both more specific and more flexible in its requirements.

4.2. OECD Compatibility

An OECD Test Guideline for Density (OECD Expert Group Physical chemistry, 1978) lists many of the same standard techniques. We believe the present document is entirely compatible with the OECD standard, but to assure adequate selection of applicable techniques, the TSCA Standards specify which protocol is to be used for specific types of materials.

C. IRLG Compatibility

The IRLG has not proposed a standard for density. We expect to submit this standard to IRLG as a proposed standard.

among three options, all of which are used to some extent in nationally and internationally standardized tests. The 20°C reference temperature is adopted by CIPAC (Raw 1970), and this provides the justification for its use. However, as explained in Section III, there is substantial justification for allowing other reporting temperatures. In summary, the TSCA protocol is compatible with the FIFRA guidelines (and with proposed revisions) but is somewhat broader in scope and both more specific and more flexible in its requirements.

4.2. OECD Compatibility

An OECD Test Guideline for Density (OECD Expert Group Physical chemistry, 1978) lists many of the same standard techniques. We believe the present document is entirely compatible with the OECD standard, but to assure adequate selection of applicable techniques, the TSCA Standards specify which protocol is to be used for specific types of materials.

4.3. IRLG Compatibility

The IRLG has not proposed a standard for density. We expect to submit this standard to IRLG as a proposed standard.

5. RESPONSES TO COMMENTS RECEIVED ON PROPOSED GUIDELINES FOR PREMANUFACTURE TESTING (USEPA 1979).

5.1. General

Following publication of the Guidelines a reconsideration of the variety of chemical materials requiring testing and of the availability of standardized procedures for such testing brought us to the conclusion that the techniques proposed then were inadequate to cover the multiplicity of identifiable situations that would arise. As a result this proposed Test Standard goes into considerably more detail with respect to the characteristics of materials that influence the selection of a technique, and it specifies appropriate techniques for handling materials with these characteristics. The requirement for the temperature at which the test is to be carried out was also changed to allow selection of one of three options.

5.2. Responses to specific comments

In response to the Proposed Guidelines, comments dealing specifically with density/relative density were received from six respondents.

5.2.1. General Approval

The largest group of responders who commented on the test expressed general approval of the test requirement, on the basis

that the test can be and routinely is made on materials for use in marketing and handling, and can be done with the accuracy required here.

5.2.2. Marginal Utility

One comment was that the test result is useful in understanding the general nature of a specific chemical but is of marginal utility in determining its environmental effects. The Agency position is that the information is needed to evaluate environmental fate, and therefore plays an important part in risk assessment. The usefulness in determining environmental fate is discussed in Section I of this document.

5.2.3. Limitations on Materials to Which the Test Should Be Applied.

One respondent stated that testing would be unneccessary if the relative density had already been determined to be less than 0.97 or more than 1.03 in the temperature range between 10 and 30°C, or if the solubility in water is 10 percent or more.

The criteria as to whether testing is to be required under a Section 4 Test Rule are somewhat different from the criteria for testing under Section 5 Premanufacture Test Guidelines. It might indeed be decided that for a particular chemical which available information put within the bounds suggested by the respondent, no test requirement would be made. On the other hand, for other material testing may be required. An example might be a dense

material that may be expected to get into flowing water where it would be transported in turbulent suspension, as sand is transported. For such material, knowledge of density is required to apply equations of turbulent transport to estimate where and how the material will move in the environment.

5.2.4. Limitations on Accuracy and Care Required

One responder suggested that special care cited in the Proposed Guideline is not required if the accuracy specified for the test is only ± 0.1 g cm^{-3}. This is an editorial comment and an appropriate change has been made.

5.2.5. Additional Test Procedures Suggested

Individual respondents suggested as alternatives: (1) a microsyringe method developed by Burroughs and Goodrich (1974); (2) Volumetric flask (Class A) (NBS 1959); (3) Simpler techniques approved by the Association of Official Analytical Chemists (AOAC 1975).

The first response to these suggestions is that the number of specified acceptable test procedures in this Test Standard is substantially larger than the number in the Guidelines (USEPA 1979), but that EPA still has not attempted to include all of the available tests that may be acceptable. While some techniques that might be acceptable are not included here, EPA feels that it has included a broad range of tests that are widely accepted and in common use. Many laboratories use the techniques, and their

applicability, limitations, and special precautions necessary will not present a hardship or increase the cost of testing. The exclusions of other methods are considered to be of minor impact in view of the large number of acceptable techniques and the long period of time over which they have been developed.

The microsyringe technique (Burroughs and Goodrich 1974) is not included, principally because no evidence was found that it has been found generally useful enough to be widely adopted. It seems most useful for rare or research chemicals that are available only in very small amounts. This criterion does not apply to chemicals subject to control under TSCA. The use of a calibrated volumetric flask is included in the specified array of acceptable techniques for the present Test Standard. The standards of AOAC (1975) included several techniques, all of which are also found in the specified array of techniques, though not specifically identified as AOAC standards.

5.2.6. Temperature of the test

One respondent suggested that the temperature range (20 ± 3)°C proposed by EPA (1979) be modified to (25 ± 3)°C in order to include most common laboratory temperatures without the need for a thermostated water bath or laboratory air-conditioning. The present test standard specifies one of three temperatures, 15°C, 20°C, or 25°C, discussed and justified in section II.B.(1) of this document.

6. REFERENCES

ASTM. 1978*. American Society for Testing and Materials. Annual book of ASTM Standards, books 1-47. Philadelphia, PA: American Society for Testing and Materials.

AOAC. 1975. Association of Official Analytical Chemists. Official methods of analysis of the association of official analytical chemists, 12th ed. Washington, DC: Association of Official Analytical Chemists.

Bauer N, Lewin SZ. 1971. The determination of density. In: Weissberger A, Rossiter BW, eds. Techniques of chemistry. New York: Wiley-Interscience, Vol. I, Part IV, Chapter 2.

Burroughs VE, Goodrich CP. 1974. Rapid method for determining densities of liquids using microsyringes. Anal. Chem. 46: 1614.

Enviro Control, Inc. 1980. Cost analysis methodology and protocol estimates: environmental standards. Rockville, MD: Enviro Control, Inc., Borriston Laboratories, Inc.

IUPA. 1975. International Union of Pure and Applied Chemistry. Commission on Physicochemical Measurements and Standards. Herrington EFG, ed. Recommended reference materials for the realization of physicochemical properties. Pure Appl. Chem. 40:393-472.

IUPAC. 1976. International Union of Pure and Applied Chemistry, Commission on Physicochemical Measurements and Standards. (a)Herrington EFG, ed. Recommended reference materials for the realization of physicochemical properties. Brown J, Lane JE, collators. Section: Density. Pure Appl. Chem. 45:1-9. (b) Physicochemical measurements: Catalogue of reference materials from national laboratories. Pure Appl. Chem. 48:503-515.

IUPAC. 1972. International Union of Pure and Applied Chemistry, Commission on Thermodynamics and Thermochemistry. A guide to procedures for the publication of thermodynamic data. Pure Appl. Chem. 29:397-407.

*As a matter of good practice, the most recent available edition that contains the desired Standard should be used.

McGlashan ML, Paul MA, Whiffen DH, eds. 1979. Manual of Units, 2nd revised 1973 edition. New York: Pergamon Press. See also: Pure Appl. Chem. 51:1-41.

NBS. 1959. National Bureau of Standards. Capacity tolerances for volumetric flasks. National Bureau Standards Circular 602. Washington, DC: U.S. Government Printing Office.

OECD. 1978. Organization for Economic Cooperation and Development Expert Group Physical Chemistry. Test guidelines for density. Draft October 11, Berlin, FRG: Umweltbundesamt.

Raw, GR, ed. 1970. CIPAC handbook, Ashworth RdeB, Henriet J, Lovett JF, compilers. Volume I. Analysis of technical and formulated pesticides. Herpender, Herts. UK. Collaborative International Pesticides Council, Ltd. (Obtain from National Agricultural Chemicals Association, Washington DC). Method MT-3. pp. 830-839.

USEPA. 1978. U.S. Environmental Protection Agency. Proposed guidelines for registering pesticides in the United States. Fed. Regist., July 10, 1978, 43 29695-29741.

USEPA. 1979. U.S. Environmental Protection Agency. Toxic substances control. Discussion of premanufacture testing Policy and technical issues; request for comment, Fed. Regist. March 16, 1979, 44 16240-16292.

Weast RF, ed. 1978-79. Handbook of chemistry and physics, 59th edition. Cincinnati, OH: CRC Publishing Co.

MELTING TEMPERATURES

STANDARD FOR TEST DATA DEVELOPMENT
PROPOSED RULE, SECTION 4, TSCA

Refers to

Part 772 — Standards for Development of Test Data
Subpart 1. Physical, Chemical and Environmental
Persistance Characteristics
Section 772.122-2 Melting Temperatures

October 1980

CONTENTS

Page

INTRODUCTION AND SUMMARY OF THE STANDARD..... 1

1. NEED FOR THE TEST..... 3

2. SCIENTIFIC ASPECTS..... 4

2.1. Melting Behavior of Materials..... 4

2.2. Test Material..... 5

2.3. Good Laboratory Practices..... 6

2.4. Test Conditions..... 7

2.4.1. Thermometers..... 7

2.4.2. Apparatus..... 8

2.4.3. Reference Materials..... 8

2.5. Test Procedures..... 9

2.5.1. Visual Technique..... 9

2.5.2. Preliminary Examination..... 9

2.5.3. Selection of a Standard Technique..... 10

2.5.4. Precise Study..... 12

2.6. Test Data Required..... 12

CONTENTS (continued)

3. ECONOMIC ASPECTS..... 14

4. TSCA/FIFRA COMPATIBILITY..... 15

5. DISCUSSION OF COMMENTS SUBMITTED IN RESPONSE TO PROPOSED GUIDELINES FOR PREMANUFACTURE TESTING..... 17

5.1 General..... 17

5.2 Specific Comments..... 17

6. REFERENCES..... 19

INTRODUCTION AND SUMMARY OF THE STANDARD

The Support Document to this test summarizes the scientific basis of the melting temperature test and its applicability in the assessments of chemicals.

The proposed test standard, when applied through a testing rule will require sponsors to determine melting temperature of the test chemical, or the range of temperature over which it melts, within the range from -30°C to +250°C. The standard is written in a general way to permit selection of a technique appropriate to the particular chemical from a variety of available techniques. The techniques proposed are limited to visual techniques in which the actual melting process is observed.

A preliminary examination of the chemical is required in order to provide information for the selection of an appropriate technique. Criteria include: (1) decomposition on melting; (2) volatility; (3) reactivity with air or with possible apparatus material; (4) lack of sharpness of melting; and (5) melting outside the range specified.

A variety of standardized techniques are stipulated for the test, but at the option of the sponsor techniques of greater accuracy than is required by the Agency are allowed. Under this standard, persons subject to TSCA physical properties test rules would develop and submit to the Agency the following kinds of information.

(1) Information and results obtained from the test, including a numerical value of the melting temperature; range of melting temperatures, or pour point; and a statement of special characteristics of the substances that influenced the selection of technique such as volatility, chemical decomposition, or range of softening temperature;

(2) Information relating to the technique used; justification for the technique selection, if a technique was used that is not specified in the standard; and

(3) Information relating to good laboratory practices, such as nature and purity of the test substance, qualifications of personnel conducting the measurement and of the laboratory in which the test was made, and quality control practice in the laboratory.

1. NEED FOR THE TEST

The normal melting temperature describes quantitatively the conditions under which a chemical substance or mixture will be solid or fluid under ambient pressure. It therefore provides information on the physical form in which the chemical will be found when in the free state.

Information on the physical form of a chemical in the environment, as solid or liquid, is important in order to assess the movement and fate of the chemical and the human and environmental exposure to it. Physical form determines whether the chemical will stand in place or flow away from spills. When combined with density information and solubility information it determines how the chemical will move in a waterway. When combined with other information such as volatility and particle size, it determines the distribution resulting from dispersal of the material. All of this information, when assessed together, is used to determine what human populations, classes of organisms, or sectors of the environment will be affected.

Knowledge of physical form also is of value in assessing the mode of interaction and speed of interaction at interfaces, which is strongly dependent on whether a substance is solid or liquid. A liquid, for instance may wet the skin and cause an inflammation or absorption much more rapidly than a solid.

2. SCIENTIFIC ASPECTS

2.1. Melting Behavior of Materials

Melting temperature is the temperature at which the change from solid to liquid takes place. The term "melting point" is often used instead.

The melting temperature of a material is affected by its composition. Impurities added to a pure substance cause a lowering of the melting temperature if the impurities are soluble in the liquid and if the mixed substances segregate on solidifying. If the mixed substances form a liquid solution and do not segregate on solidifying, the melting temperature will usually be a weighted average of the melting temperatures of the substances in the mixture.

Other materials that form glasses have special melting behavior because of complex intermolecular forces, and the distinction between solid and liquid is not clear cut.

Thus, in practice, many materials, particularly those that are mixtures or form glasses, do not melt sharply at a well defined temperature, but rather soften and liquefy over a range of temperatures. Under some circumstances the melting temperature is lowered many kelvins (degrees Celsius) by impurities. For these materials other terms are used, including pour point, congealing temperature, drop melting temperature, initial and final melting temeprature, or melting range. Such

parameters are included in this test standard and are defined in the standard.

Definitions related to normal melting temperature are given to allow for more precision of statement in describing the results of tests on impure materials and materials that do not have sharp melting temperatures. They also are useful in describing tests that inherently do not strictly determine the true melting temperature, but only an approximation to it. These terms are in common practice in test laboratories and test standards.

The term "normal" is often used to specify that the phase change occurs at 1 atm pressure. Because the effect of pressure changes is rather small, we do not emphasize the use of this term. The term "melting temperature" is used instead of "melting point" to specify that it is the temperature rather than pressure or volume that is of interest, and to emphasize the fact that "point" refers to a "point" on a phase diagram (i.e., an invariant), while the melting temperature is a variable depending on pressure and composition.

2.2. Test Material

The type or grade of material to be tested is not specified in this Standard, but will be specified in Test Rules that incorporate the Standard. The following discussion relates to the choice of material to be tested.

The normal melting temperature of a pure material is one means of characterizing the substance and usually is considered a worthwhile standard reference datum.

The technical grade of substance is often a mixture of the substance with other substances that are not easily separated and do not interfere with the technological applications of the material. Because of the phase behavior of materials briefly outlined in earlier parts of this Standard, the technical grade material will usually have a melting temperature different from that of the pure substance and melt over a range of temperature, rather than sharply at a single temperature. These effects may amount to several kelvins, and may not be the same from one batch of technical material to the next. The technical grade is the material that will normally be found in commerce, and the temperature differences that will be encountered are within the range of fluctuations that occur periodically in the environment.

2.3. Good Laboratory Practices

Laboratories that do not have proper quality control sometimes produce data that are less than adequate for supporting sound analyses and decisions. Good Laboratory Practice Standards, cited in section (c) of the test standard, are designed to ensure that all data generated in response to Test Rules will be valid. For further discussion of the details contained in Good Laboratory Practice Standards see the Preamble

for Good Laboratory Practice Standards for Physical, Chemical, Persistence, and Ecological Effects Testing (Section 772.110-2) proposed in the same issue of the Federal Register in which this proposed Standard for density is proposed.

2.4. Test Conditions

2.4.1. Thermometers

A good thermometer is the most important item of equipment needed for measuring melting temperature. Specifications for all the thermometers needed for the techniques listed are given by ASTM.

Thermometers frequently are inaccurate because of errors in manufacture or accidents which damage them, such as by separation of the mercury or fluid column. They, therefore, invariably need to be calibrated if they are to be accurate indicators of temperature.

The U.S. temperature scale is maintained at the National Bureau of Standards, and measurement accuracy is propagated throughout the nation by calibrations of thermometers originating at the National Bureau of Standards. Traceability to the National Bureau of Standards may be by comparison of thermometers of which one was calibrated at the National Bureau of Standards, or by measurement of the melting temperature of a substance certified for melting temperature at the National Bureau of Standards, or by a longer sequence of comparisions. The

comparison can be done by the manufacturer or by the laboratory doing the testing. If this traceability is not available for the thermometer being used, there is no way to verify readings that are taken during the course of testing.

2.4.2. Apparatus

See the discussion of test procedures (section 2.5. of this document) for a description of how specific test procedures were selected and adopted from among standardized methods. Published protocols for the standardized methods generally include descriptions of specific pieces of equipment or apparatus and accessories required for the test. The equipment, as specified in the protocol, is considered to be an integral part of the standardized test, and therefore is required by EPA as one of the conditions of this Standard.

2.4.3. Reference Materials

See the discussion of the use of reference materials for the calibration of thermometers in a previous paragraph in this same section (2.4.) of this support document. In addition, the reference materials should be available and should be used for the calibration of the total apparatus and procedure used. Some of the standardized protocols adopted in this standard specifically require a calibration step with a standard reference material. Several recommended materials are listed in the standard, and should be selected by the analyst according to the

method used and the type of material being tested. The standard also allows for the selection of other materials as the analyst may judge necessary.

2.5. Test Procedures

2.5.1. Visual Technique

The criterion of visual evidence of melting is necessary to establish that the transition reported is actually that between solid and liquid. This is because of the pronounced difference in the behavior of solids and liquids in dispersion, in rate of reaction, and in kind of surface interaction with other materials.

The list of acceptable techniques could be expanded by including nonvisual techniques such as differential scanning calorimetry (DSC). This would be permissible only after evidence was obtained in preliminary visual examination that the melting occurred in a general temperature range within which a single DSC-observed transition could be indentified. This seemed to constitute a more complex test than those adopted and was thus rejected.

2.5.2. Preliminary Examination

The purpose of the preliminary examination required by this Test Standard (Section (e)(2)) is to identify special features of the test material that would invalidate a test of melting

temperatures and to guide the selection of a suitable technique. The following section describes how the information from the preliminary examination is applied in the selection of one of the standard techniques referenced in the Test Standard.

2.5.3. Selection of a Standard Technique

Section (e)(3) and Table 1 of the Test Standard require the use of certain standardized protocols to be selected according to the information determined from the preliminary examination.

In selecting the many standardized techniques listed in Table 1, EPA reviewed many published protocols, and determined that those selected were capable of producing good and reliable data for the broad range of substances that are expected to be tested under the provisions of TSCA. These protocols are believed to be widely used for the routine determination of melting temperature, and therefore provide economical and suitable means of producing the data that EPA requires for evaluation of environmental exposure.

Paragraphs (e)(3)(i)-(iv) of the Standard specify which type of technique(s) (e.g., Thiele tube, cooling curve, pour point, etc.) is(are) to be used according to the type of melting characteristics which different types of materials have. These specifications were based on a detailed review of the protocols and their stated or EPA's judgment of their applicability. For material for which the standard gives several types of suitable techniques, the analyst is free to select the most applicable

type, and likewise, where Table 1 lists several suitable protocols for a type of test, the analyst may select the most convenient or suitable standard method to follow.

The following several paragraphs give additional detail as to how the stipulated techniques in sections (e)(3)(i)-(iv) relate to the melting characteristics of the test materials.

Softening temperatures are best determined by techniques not suitable for materials with sharp melting temperatures. If decomposition or other chemical reaction occurs on melting, the temperature observed depends strongly on the rate of heating. This is because the properties of products of the reaction affect the results. Therefore, the technique must take account of this possibility. Vaporization might cause disappearance of the sample and excessively volatile substances should be enclosed for the test.

Techniques designed for substances that melt sharply are unsuitable for substances that have an extended softening interval. This is because the techniques will show a broad range of melting temperature or will give results which have considerable uncertainty. Techniques that rely on physical appearance or movement under rigorously standardized conditions narrow the range of uncertainty for such substances.

For materials that decompose on melting, a technique needs to be used in which decomposition does not influence the results. For example, the Kofler hot bench technique effectively

divides a lot of material into small sublots, each of which is exposed to a different temperature and is separately observable. Thus, the decomposition at one temperature does not unduly influence the state of an adjacent substance which is at a slightly lower temperature. Volatilization, reaction with air, or other reactions tend to invalidate the test; hence, this limits the kind of measurement technique that can be used.

2.5.4. Precise Study

A well established research and development facility or a quality control laboratory may very accurately determine the melting temperature using procedures referenced in section (e)(4) of the Standard. The results of these more accurate tests certainly satisfy the Agency's needs, and therefore are acceptable in lieu of the standardized tests specified in section (e)(3).

2.6. Test Data Required

No melting temperature measurement is required for substances that sublime or decompose at normal pressure because the melting process does not occur under real world conditions.

Information about any nonspecified test procedure is needed to determine that acceptable data have been generated and that the testing laboratory had good reason for using a nonstandard method.

The level of accuracy required in reporting is 1 to 2°C. In general this level of accuracy is inherent in the technique, is easy to determine, and is approximately the accuracy with which ambient temperatures are known. This point is discussed further in section 5.2.(2).

Sections (f)(4)-(11) specify the observable end points which are to be reported for the different types of testing techniques. These reportable end points have been specified in the standard to assure that data from the different techniques are reported in similar manners that are suitable for use in EPA's data storage systems, are suitable for comparison from chemical to chemical, and are correctly reported for checking the performance of the method when used with standard reference materials.

3. ECONOMIC ASPECTS

A survey of three commercial laboratories to estimate costs for performing the tests outlined in this Standard found a range of $25 to $75, with a "best estimate" of $50. A cost estimate also was made by separating the protocol into components and estimating the cost of each component, including direct labor cost, overhead cost, other direct costs, general and administrative costs, and profit or fee. The protocol best estimate of cost was $83, with an estimated range of $42 to $125 based on ± 50 percent of the best estimate. An analysis of the discrepancy between the protocol estimate and the survey pointed out that the nature of the chemical to be analyzed has an impact on the cost of analysis. The actual melting temperature of the compound has an influence on cost. For example, a compound melting at 250°C will take longer to analyze than one with a lower melting point. Separate methods also are required for compounds that decompose or melt, compounds that show extended softening, and compounds that react or volatilize. Differences in the methods will cause variations in the cost.

The above cost estimates were made assuming that all the requirements of Good Laboratory Practice Standards, as specified in section (c) of the melting temperature standard, are being satisfied. Details of the cost estimate are in a report by Enviro Control, Inc. (1980).

4. TSCA/FIFRA/IRLG/OECD COMPATIBILITY

In Pesticides Guidelines (USEPA 1978) Section 163.61 8(c)(3) a melting temperature or melting range in °C is required for a solid technical chemical unless the technical chemical sublimes or decomposes. This criterion is consistent with the present Test Standard.

In Pesticides Guidelines (USEPA 1978) Section 163.61, Appendix to Product Chemistry calls for CIPAC test MT 2. This is one of the standard tests using the Thiele tube to which we refer. Any data acceptable under Pesticide Guildlines would therefore be acceptable under this Test Standard, provided that the test substance was appropriate and did not require use of one of the specialized methods specified in Section (e)(3)(i), (ii), or (iii) of the Standard.

There is a difference in the scope of requirements for FIFRA and TSCA in that the FIFRA applies solely to pesticides, which are a more uniform chemical group than those to which TSCA applies. The CIPAC test is an adequate test for many pesticides; it is not adequate for the whole range of chemicals to which TSCA applies. Therefore, this Test Standard appropriately includes a broader range of tests than Pesticide Guidelines because the greater range of chemical types requires a greater range of techniques.

The OECD (1979) has issued a Draft Test Guideline for Melting Point/Melting Range. This OECD draft guideline includes

a variety of techniques, allowing for making the determination on materials with various melting characteristics, and in this way it is comparable to the proposed TSCA standard. The OECD draft is considerably longer and gives many experimental details and other parameters that are included in the proposed TSCA standard only by reference to ASTM or other standards. A technique-by-technique comparison shows that the OECD guideline and the EPA proposed standard have scopes which seem to differ principally in the inclusion in the EPA standard of provision for materials that soften and melt only over an extended range, and giving a drop melting temperature or pour point. The proposed TSCA Standard does not discuss automated methods, whereas the OECD standard discusses the applicability of automation to several, and includes one (photocell detection) that is only offered as an automated technique. From EPA's point of view, automation is a peripheral factor because TSCA tests fundamentally are not production-line processes. The EPA standard outlines to a much greater degree how the testing of a particular substance is to be approached but avoids many of the details found in the OECD standard.

5. DISCUSSION OF COMMENTS SUBMITTED IN RESPONSE TO PROPOSED GUIDELINES FOR PREMANUFACTURE TESTING (USEPA 1979).

5.1. General

Following publication of EPA's Proposed Guidelines for Premanufacture Testing, the Agency reconsidered the variety of chemical materials requiring testing under Section 4 of TSCA and the availability of standardized procedures for determining melting temperature. EPA concluded that the techniques originally proposed were inadequate to cover the multiplicity of identifiable situations that would arise. As a result, this proposed Test Standard gives considerably more detail with respect to the special characteristics of materials that influence the selection of a technique, and it specifies appropriate techniques for handling materials with these characteristics.

5.2. Specific Comments

In response to the Proposed Guidelines (USEPA 1979) comments referring specifically to melting temperature were received from 10 responders. Their comments may be grouped in five categories:

(1) General concurrence. Three responders expressed general approval of the tests and the reference to standardized procedures such as those of ASTM.

(2) Precise measurements are not needed. We concur. However, we do not believe the requirement of accuracy can be relaxed so far as to allow a range of 12°C as suggested by one responder. While environmental

temperatures sometimes fluctuate daily by this amount and seasonal changes are even larger, the physical state of the chemical will also fluctuate if it happens to have a melting temperature within the daily or seasonal fluctuation range. Thus, for a reasonably good assessment of the state and manner of movement of the chemical in a particular environment the temperature of melting should be known with approximately the same certainty as the temperature of the environment is known, 1 to 2°C.

(3) Measurement is redundant and should be omitted. Three responders made this argument on the basis that melting and boiling temperature were lumped together for discussion in the Proposed Guidelines. They argued that vapor pressure measurement gives the essential information contained in a boiling temperature measurement. An examination of the argument reveals that it must have been directed toward the boiling temperature alone and is invalid when applied to the melting temperature. The boiling temperature is a rapidly varying function of pressure, while melting temperature is a slowly varying function and futhermore, melting temperature cannot be derived from the vapor pressure alone.

(4) Measurement as described is inapplicable to polymers. We concur with the two responders making the point that the Proposed Guideline (USEPA 1979) was inadequate in this respect. We believe the present proposed Test Standard with additional protocols is versatile enough to provide for the necessary softening or melting temperatures that are useful for characterizing polymers.

(5) Refer to specific test techniques. One responder indicated that their laboratory regularly used differential thermal analysis (d.t.a.) for this test and that a test costs $2500. The d.t.a. method was in the Proposed Guidelines (USEPA 1979), but d.t.a. has been omitted from the current Test Standard because of excessive cost. The fact that the responder quotes a cost of $2500, compared to costs for traditional melting temperature tests in the range of less than $125 (Section III) supports our contention that the d.t.a./d.s.c. technique is more elaborate and costly than the techniques specified. At the sponsor's option, however, the d.t.a./d.s.c. techniques still could be used under provisions of section (e)(4).

6. REFERENCES

Enviro Control, Inc. 1980. Cost analysis methodology and protocol estimates: environmental standards. Rockville, MD. Enviro Control, Inc., Borriston Laboratories, Inc. April 28.

OECD. 1979. Organization for Economic Cooperation and Development. Draft test guideline for melting point/melting range. Berlin: Umweltbundesamt. June 20.

USEPA. 1979. U.S. Environmental Protection Agency. 1979. Toxic substances control. Discussion of premanufacture testing policy and technical issues; request for comment. Fed. Regist., March 16, 1979, 44 16240-16292.

USEPA. 1978. U.S. Environmental Protection Agency. Proposed guidelines for registering pesticides in the United States. Fed. Regist., 1978, 43 29696 - 29713.

VAPOR PRESSURE

STANDARD FOR TEST DATA DEVELOPMENT
PROPOSED RULE, SECTION 4, TSCA

Refers to

Part 772 — Standards for Development of Test Data
Subpart L. Physical, Chemical and Environmental
Persistence Characteristics
Section 772.122-3 Vapor Pressure

October 1980

CONTENTS

		PAGE
1.	Need for the Test	1
2.	Scientific Aspects	5
	2.1. Test methods	5
	2.2. Test procedures	9
	2.2.1. Temperature of the test	9
	2.2.2. Sorbent for gas saturation procedure	10
	2.2.3. Gas flow rates in gas saturation procedure	11
	2.2.4. Calculations of vapor pressure in gas saturation procedure	11
	2.3. Test data required	12
3.	Economic Aspects	14
4.	Replies to comments on March 16, 1979 document	16
5.	TSCA/FIFRA Compatibility	20
6.	TSCA/OECD Compatibility	21
7.	References	22

1. NEED FOR THE TEST

The vapor pressure of a chemical is an important parameter in determining the environmental fate of the chemical.

The atmosphere is a major route for the widespread distribution of chemicals. There are several ways by which chemicals may become airborne and subsequently be transported by wind currents. Airborne solids and foamy emulsions are commonly observed, but these are not believed to be major factors in atmospheric transport because they involve particulate matter which settles out in a relatively short time (Seiber et al. 1975). Aerosols, from spray applications, manufacturing and formulation sites, and aerated waste treatment systems may constitute more important sources of chemical for air transport since very small droplets (5 micrometers or less in diameter) may be formed and carried considerable distances (Edwards 1973). However, it appears that volatilization from land and water surfaces is the most important source of material for airborne transport (Hartley 1969, Lichtenstein 1971, MacKay and Wolkoff 1973, Seiber et al. 1975).

Volatilization is the evaporative loss of a chemical compound. Volatilization rates are dependent on the vapor pressure of the chemical and the environmental factors which influence diffusion from the evaporative surface. Harper et al. (1976, p. 236) noted that "volatilization is probably the single largest means by which pesticides are lost and transported over wide areas and into bodies of water far from the application

location." The airborne vapors of a hazardous chemical may present a threat to plant and animal life exposed to those vapors, not only in the area of chemical release but also at sites remote from the volatilization site. This occurs when vapors are removed from the air, primarily by precipitation with rain or snow.

Volatilization rates are related to vapor pressure, which varies with temperature. However, volatilization from soil or water is influenced by other environmental conditions and the effective vapor pressure may be considerably lower than the potential vapor pressure. Nevertheless, vapor pressure is the one common factor governing the tendency of a compound to volatilize.

According to kinetic theory there is a continuous flight of molecules from the surface of a liquid or solid into the free space above it. At the same time vapor molecules return to the surface at a rate depending on the concentration of the vapor. If there is no removal of vapor from the surface (for example, by air currents), equilibrium will be established where the rate of vaporization is exactly equal to the rate of condensation. The pressure exerted by the equilibrium vapor is known as the vapor pressure (Daniels and Alberty 1955) and is dependent upon temperature.

Knowledge of the vapor pressure of a compound allows the ranking of a chemical as relatively nonvolatile, highly volatile, or of some intermediate volatility. When vapor pressure data are combined with solubility data to calculate Henry's Law constants,

as described by MacKay and Leinonen (1975) and Dilling (1977), rates of the evaporation of dissolved chemicals from water can be estimated.

Evaporation from an exposed surface will depend upon other factors such as wind speed (which reduces the vapor density above the surface) and adsorption (which may act to hold the substance on the surface). Volatilization from aqueous systems depends also on the solubility of the compound and its movement to the water surface. In soils the rate of volatilization of a chemical will depend upon such factors as adsorption on soil, solubility in soil water, and on the amount of soil water and its rate of evaporation. Volatilization from soils can become a diffusion controlled process as mass transfer to the soil surface is reduced by low water evaporation due to high humidity or to the lack of soil water in a dry soil (Bailey et al. 1974).

Chemicals that have relatively low vapor pressures and that sorb readily to solids or dissolve readily in water are not likely to vaporize significantly at ambient temperatures. For that reason, airborne transport is not a major transport mechanism for these chemicals and assessment of them should be focused on their chemical fate and environmental effects in soils, sediments, and water. However, chemicals with high vapor pressures or with relatively low water solubility and low adsorptivity to solids are less likely to reside only in soils, sediments or water, since volatilization can be a potentially significant factor in their environmental transport. Chemicals that are

gases at ambient temperatures and that have low water solubility and low adsorptive tendencies will be transported to a significant degree in the atmosphere and are prime candidates for photolysis and for involvement in adverse atmospheric effects such as smog formation or stratospheric alterations. Furthermore, effects testing of those chemicals should also focus on inhalation and surface contact as potential routes for direct exposure.

An understanding of how a chemical is likely to partition among the various environmental media (air, water, soil, and sediment) is needed in judging whether or not a chemical will be subject to various transformation possibilities, such as oxidation by hydroxyl ion or ozone in the atmosphere. Vapor pressure data can influence decisions on whether or not it is appropriate to conduct photolysis, adsorption/desorption, partition coefficient, and certain biodegradation tests. Vapor pressure data are an important consideration in the design of other fate and effects tests, for example in preventing or accounting for the loss of volatile materials during the course of the test. Clearly, a knowledge of vapor pressure combined with information on water solubility and adsorptive tendencies is necessary in predicting environmental transport and in providing guidance as to which persistence and effects tests need to be considered and how those tests should be designed.

2. SCIENTIFIC ASPECTS

2.1. Test Methods

A procedure for measuring the vapor pressure of materials released to the natural environment ideally would cover a range of vapor pressure values, at ambient temperatures, of about 10^{-5} Pa to 10^{5} Pa (approximately 10^{-7} to 760 Torr). Because no single procedure can cover this range, two different procedures are described, each suited for a different part of the range. The isoteniscope procedure (ASTM 1978) is for pure liquids with vapor pressures from 0.1 to 100k Pa. For vapor pressures of 10^{-5} to 10^{3} Pa, a gas saturation procedure is to be used. The Knudsen effusion procedure (Thomson and Douslin 1971) may be used for low vapor pressure values.

Each of the tests must be performed under conditions of normal laboratory room temperatures in order to allow for careful control of the temperatures in thermostated baths or chambers containing the test apparatus.

The isoteniscope procedure uses a standardized technique that was developed to measure the vapor pressure of certain liquid hydrocarbons. It is applicable to pure liquids with vapor pressures of 0.1 kPa (0.75 Torr) or more at ambient temperatures. The sample is purified within the equipment by removing dissolved and adsorbed gases until the measured vapor pressure is constant. This process is called "degassing." The procedures do

not remove higher boiling impurities, decomposition products, or compounds that boil close to or form azeotropes with the material under test. If compounds that boil close to or form azeotropes with the test material are present, it is necessary to remove the interfering compounds and use pure test material. Impurities more volatile than the sample will tend to increase the observed vapor pressure above its true value but the purification steps will tend to remove these impurities. Soluble, nonvolatile impurities will decrease the apparent vapor pressure. However, because the isoteniscope procedure is a static, fixed-volume method in which an insignificant fraction of the liquid sample is vaporized, it is subject to only slight error for samples containing nonvolatile impurities. That is, the nonvolatile impurities will not be concentrated due to vaporization of the sample.

Gas saturation (or transpiration) procedures use a current of inert gas passed through or over the test material slowly enough to ensure saturation and subsequent analysis of either the loss of material or the amount (and sometimes kind) of vapor generated (Bellar and Lichtenberg 1974, Thomson and Douslin 1971).

The gas saturation procedures have been described by Spencer and Cliath (1969). Results are easy to obtain and can be quite precise. The same procedures also can be used to study volatilization from laboratory scale environmental simulations. Vapor pressure is computed on the assumption that the total

pressure of a mixture of gases is equal to the sum of the pressures of the separate or component gases. The partial pressure of the vapor under study can be calculated from the total gas volume and the weight of the material vaporized. If v is the volume which contains w grams of the vaporized material having a molecular weight M, and if p is the pressure of the vapor in equilibrium at temperature T (K), then the vapor pressure, p, of the sample is calculated by

$$p = (w/M)(RT/v)$$

where R is the gas constant (8.31 Pa $m^3 mol^{-1} K^{-1}$) when the pressure is in pascals (Pa) and the volume in cubic meters. As noted by Spencer and Cliath (1970), direct vapor pressure measurements by gas saturation techniques are more directly related to the volatilization of chemicals than are other techniques.

In an effort to improve upon the procedures described by Spencer and Cliath and to determine the applicability of the gas saturation method to a wide variety of chemical types and structures, EPA is sponsoring research and development work at SRI International (EPA Contract No. 68-01-5117). The procedures described in Appendix A are those developed under that contract and currently being evaluated with a wide variety of chemicals of differing structure and vapor pressures.

In addition to the above methods, other procedures have been described for the measurement of vapor pressure (Daniels et al. 1956, Glasstone 1946, Nesmeyanou 1963, Thomson and Douslin 1971). These include boiling point procedures, effusion techniques, and many highly specialized techniques that are restricted to the determination of very precise vapor pressure values or to the measurement of vapor pressures of specific kinds of materials. These highly specialized methods were rejected by EPA because they do not have general applicability to either a wide variety of chemicals or a relatively broad range of vapor pressure values at ambient temperatures.

EPA rejected boiling point procedures, such as that using Ramsey and Young apparatus, because the accuracy is very poor below 10^3 Pa (Thomson and Douslin 1971) and because boiling point methods provide inaccurate estimates of the vapor pressures at ambient temperatures if there is a change of state or a transition temperature between the boiling temperature and ambient temperatures.

Effusion techniques, particularly those employing the Knudsen effusion apparatus, are used to measure vapor pressures from about 10^{-5} to 1 Pa and have provided some good data (Hamaker and Kerlinger 1969). Those procedures were rejected because they require working with systems under vacuum and because it is necessary to saturate the capsule space with vapor during the measurement periods. The lack of equilibrium saturation has been postulated as a reason for inaccurate published vapor pressures data (Spencer and Cliath 1970). However, it must be recognized

that there are laboratories which have employed Knudsen effusion techniques successfully and which have considerable experience with the method, especially for determining very low vapor pressure values, such as 10^{-5} to 10^{-3} Pa. For such laboratories, the Knudsen effusion methods are a satisfactory alternative to the gas saturation method in the determination of low vapor pressure values. However, it seems reasonable to require that the laboratory using effusion methods supply documentation to substantiate successful utilization of the effusion procedures with other compounds.

2.2. Test Procedures

2.2.1. Temperature

The test procedures generally require a thermostated bath or test chamber temperature of 25 ± 0.5° C. Laboratories should be able to carry out vapor pressure measurements without the need for elaborate temperature control devices. Control of the bath or chamber to ±0.5°C will permit substantial confidence in the data without requiring unnecessarily costly apparatus.

The International Union of Pure and Applied Chemistry has for many years (IUPAC 1972) recommended the reporting of physical-chemical properties measurements at the temperature of 25°C. A temperature of 25°C is slightly above most laboratory room temperatures and this allows for convenient adjustment and maintenance of constant temperature baths and enclosures.

Because of the nature of the isoteniscope procedure, it is necessary in that test to conduct some measurements at temperatures above and/or below 25°C in order to determine whether the sample needs further degassing. Also, for some chemicals, it may be necessary to require vapor pressure data at temperatures other than 25°C and, in such cases, this requirement will be noted in Test Rules for specific chemicals. Examples of when this requirement may be applicable include situations where there is evidence that the vapor pressure may change significantly with relatively small changes in ambient temperature or when the boiling temperature for a chemical is at an ambient temperature below 25°C.

2.2.2. Sorbent for Gas Saturation Procedure

The choice of sorbent and desorption solvent are dictated by the nature of the compound being evaluated. Charcoal sorbent is inexpensive and may be desorbed with carbon disulfide, a convenient solvent for use with flame ionization detector. Many compounds, however, do not desorb efficiently from charcoal and more expensive sorbents, such as Tenax GC and XAD-2, must be used. The desorption efficiency of a particular compound from a sorbent with a solvent is defined as the weight of the compound which can be recovered from the sorbent divided by the weight of the compound originally adsorbed. It must be measured for every combination of sample, sorbent, and solvent used. Desorption efficiency may vary with concentration, so it is important to

measure it at or near the concentration of the actual sample. It is sometimes necessary to interpolate between two measured efficiencies.

2.2.3. Gas Flow Rates in Gas Saturation Procedure

Accurate control of gas flow rates is essential to assure that a known volume of carrier gas is passed through the system. Very long sampling times are required for compounds with low vapor pressures, and it is difficult to control very low flow rates for very long times. It is necessary to use fine needle valves to control the flow rates and to measure the flow rates frequently during the test period in order to make corrections for variation which can occur, e.g. due to changes in atmospheric pressure.

2.2.4. Calculations of Vapor Pressure in the Gas Saturation Procedure

The calculation of vapor pressure is straightforward. The weight of the sample desorbed from a sorbent section is divided by the desorption efficiency to give the weight of the sample collected by the sorbent trap. With the volume of carrier gas calculated from the flow rate, the ideal gas law is used to calculate the vapor pressure of the sample. To assure that the carrier gas is indeed saturated with the compound vapor, each compound is sampled at three different gas flow rates. If the vapor pressure calculated shows no dependence on flow rate, then

the gas is assumed to be saturated. The method also assumes that there are no interactions between vaporized sample and the carrier gas and that the molecular weight of the vaporized sample is the same as for the sample liquid or solid. If there are any indications that these may not be valid assumptions, the vapor should be analyzed both qualitatively and quantitatively using such techniques as gas chromatography combined with mass spectrometry (Heller et al. 1975).

2.3. Test Data Required

The average calculated vapor pressure for the test material at each required test temperature must be reported. The reported data must also include the individual values from triplicate determinations and the calculated standard deviation for each average calculated vapor pressure. It might be preferable for assessment purposes to require that each vapor pressure determination be made in sufficient replication to provide a given degree of reproducibility. However, the precision attainable will vary not only with the number of replications but also with the procedure employed and the test chemical. For a given chemical, the only way to determine how many replications of a given procedure are necessary to provide vapor pressure data with some specified precision is to repeat the procedure until the data provide that precision. This may take a few or many replications and a requirement for numerous replications is not justified unless the specified precision is needed for assessment purposes with an individual chemical. The minimum requirement

EPA would impose would be a statistical analysis of vapor pressure data to provide standard deviation calculations based on triplicate determinations.

For the isoteniscope method, the vapor pressure data generated during the degassing operation, including a plot of log p vs 1/T, must be included to provide evidence of successful degassing. For the gas saturation method, the data showing that vapor pressure does not vary with flow rate must be included to provide evidence of saturation of the carrier gas with the sample vapor. The data also must include a complete description of all analytical techniques and results, a description of the sorbents and desorption solvents used and the desorption efficiency calculations.

3. ECONOMIC ASPECTS

A survey of commercial laboratories to estimate costs for performing the tests outlined in this Standard predicted a range of $64 to $193 per substance for the isoteniscope method and $185 to $554 for the gas saturation method. The "best estimate" costs, based on the survey, were $129 for the isoteniscope method and $369 for the gas saturation method. The cost estimates were made assuming that all the requirements of Good Laboratory Practice Standards (GLPs), as specified in section (c) of the Test Standard, are being satisfied. Details of the cost estimates are contained in a report by Enviro Control, Inc. (1980).

The cost estimates in the Enviro Control report do not include the costs associated with specialized analytical methodology or for analytical method development that may be required for vapor pressure analysis by the gas saturation method. It is not possible to estimate the cost of such special analytical requirements since the costs are related to the nature of the chemical to be analyzed and because such cost may be prorated among the analytical needs related to other Test Standards. Also, the nature of the chemical to be tested for vapor pressure may cause drastic variations in price when, for example, very long sampling times are required in the gas saturation method for substances with very low vapor pressures.

In reviewing the state of technology concerning this area of testing, the Agency has not identified any adequately developed tests that would be significantly less costly while providing comparable information.

4. REPLIES TO FEDERAL REGISTER OF MARCH 16

The Federal Register of March 16, 1979, (EPA 1979) presented a discussion of policy issues, alternative approaches, and test methods under consideration as guidance for premanufacture testing. Included in the test methods was a section on vapor pressure (pp. 16256-16257) which discussed the isoteniscope procedure and the gas saturation procedure as potential methods for use in developing information on new chemicals. Among the comments received in response to the discussion were several comments related to the vapor pressure methods.

Several commentors contended that a requirement for vapor pressure measurements at each of three ambient temperatures (10, 20, and 30°C) would be unnecessarily time consuming and too costly for the needs of a chemical fate assessment. There were two reasons for obtaining vapor pressure data at each of three temperatures. First, it seemed desirable to obtain vapor pressure data at representative environmental temperatures to determine if vapor pressure changed significantly over that range of temperatures. Second, the determination of vapor pressure at each of three temperatures provides a check on the validity of the data. If the vapor pressure data are determined correctly, then a plot of log p vs 1/T (where p = vapor pressure as Pa and T = test temperature as K) will produce a straight line. However, for most substances it will not be necessary to have knowledge of the vapor pressure at each of three rather closely spaced temperatures and a requirement for the determination of vapor

pressure at each of three temperatures solely as a check on the validity of the method would be unduly time consuming. Therefore, the requirement has been changed to a determination of vapor pressure at one temperature, 25°C, unless otherwise noted in a specific Test Rule.

For some chemicals there may be indications that the vapor pressure changes significantly over the range of environmental temperatures or there may be evidence that there is a change of state at an environmental temperature. Either of these circumstances may justify a requirement for vapor pressure measurements at temperatures in addition to 25°C. Note, also, that the isoteniscope method requires a determination at 25°C plus a sufficient number of other temperatures to ascertain whether or not further degassing is needed. Although the vapor pressure determinations at temperatures other than 25°C need not be made in triplicate (unless they are temperatures specified in a Test Rule), the data can serve to confirm the validity of the vapor pressure results.

Some commentors suggested that the investigator should be able to use any technique, even employing high temperatures for the vapor pressure measurement and an Antoine equation nomograph to determine the vapor pressure at ambient temperatures. The problem with this appraoch is that there may be a change of state or a transition temperature between the test temperature and ambient temperatures. If this is the case, extrapolation will yield incorrect and misleading results. The use of differential thermal analysis and thermal evolution analysis, suggested by one

commentor, involves variations on the melting point/boiling point procedures for estimating vapor pressures and are subject to the same errors described above. It may be appropriate to use such estimation techniques to provide guidance in performing the vapor pressures measurements but those estimated values for vapor pressure would not be sufficiently reliable to be used in estimating volatility using water solubility and calculating the Henry's law constant. Such predictions of volatility based on Henry's law constant will be an important consideration in evaluating the environmental fate of a chemical.

It was commented that ionizable and highly soluble compounds should be exempt from the requirements for vapor pressure data. This would be a reasonable suggestion if it could be demonstrated that the chemical in question would reside exclusively in the aqueous environment upon release to the environment. If the chemical can occur in non-aqueous environments, such as soils or on solid surfaces, its vapor pressure is needed regardless of whether or not it is ionizable or highly soluble in water.

It was stated that there should be no need to determine vapor pressure as low as 10^{-5} Pa. Unfortunately, experience has shown that compounds with very low vapor pressures can volatilize to a significant extent when low vapor pressure is combined with low water solubility. Examples include DDT and the polychlorinated biphenyls (PCBs). When dealing with the wide variety of chemical compounds which exist in the commercial chemical inventory, it is necessary to conclude a concern for those which have low vapor pressures and low water solubilities

and which may, as a result of these factors, evaporate from water bodies and become airborne contaminants.

One commentor suggested simply determining the weight loss at 1 Torr (about 10^2 Pa) and 30°C in place of a determination of vapor pressure. This simplistic sort of procedure can provide some information on the tendency of a compound to evaporate from a non-adsorptive surface, provided the vapor pressure is sufficiently high to observe results in a relatively short time, but no quantitative information is obtained which is of value in assessing environmental fate.

5. TSCA/FIFRA COMPATIBILITY

In the 1978 Proposed Pesticide Guidelines (EPA 1978) it is suggested that vapor pressure be determined by ASTM Method D-3074-72. This method measures the pressure within an aerosol can and is related to packaging. It is not applicable to environmental fate considerations. In the 1975 Proposed Guidelines (EPA 1975), it was suggested that vapor pressure should be determined by the Knudsen effusion or the gas saturation 'transpiration) procedures, both of which are discussed in this document. The FIFRA proposed guidelines have not contained any reference to a vapor pressure prcedure which would be applicable to compounds with relatively high vapor pressures, in excess, say, of 1 kPa at environmental temperatures. This may be because the FIFRA concern is primarily with compounds of low vapor pressure, although there are some pesticides which do not fit that picture, such as fumigants. Since TSCA must deal with the entire range of chemicals, the Test Standards available must include methodology suitable for chemicals with high vapor pressures, such as the isoteniscope procedure.

6. TSCA/OECD COMPATIBILITY

The Organization for Economic Cooperation and Development (OECD) has been developing a testing plan for toxic chemicals which will aid in assessing environmental fate and effects. The plan, which has not yet been published, will include recommended methodology for vapor pressure. The approach being considered by OECD is similar to that proposed by TSCA in the Federal Register discussion document of March 16, 1979, (EPA 1979). The isoteniscope method and the gas saturation method are recommended for the determination of vapor pressure at 30°C. OECD also recommends a vapor pressure balance method which seems to be similar in principle to the Knudsen effusion method and which measures the pulse generated on a sensitive balance by a vapor jet in a high vacuum. This appears to be a rather complicated procedure employing delicate apparatus. There are no literature references to its use in the OECD draft reports. The equipment and expertise may be readily available in Europe but it does not seem to be a procedure in common use in the United States. For laboratories which have the equipment and successful application experience, the Vapor Pressure Balance method may be an acceptable procedure for determining relatively low vapor pressure values, but EPA would need to have more information on the method and its use before acceptance. Finally, the OECD suggests the use of a boiling point method which EPA believes would not provide acceptable data, because of poor reproducibility and the possibilty of gross inaccuracies, as noted earlier.

7. REFERENCES

ASTM. 1978. American Society for Testing and Materials Annual book of standards, part 24. Philadelphia, PA. pp. 740-745.

Bailey GW, Swank RR, Jr., and Nicholson HP. 1974. Predicting pesticide runoff of agricultural land: a conceptual model. J. Environ. Qual. 3:95-102.

Balson EW. 1947. Studies in vapor pressure measurement, Part III--An effusion manometer sensitive to 5 x 10^{-6} mm Hg: Vapor pressure of DDT and other slightly volatile substances. Trans. Fara. Soc. 43:54-60.

Bellar TA and Lichtenberg JJ. 1974. Determining volatile organics at microgram-per-litre levels by gas chromatography. J. Am. Water Works Assn., 66:739-744.

Daniels F and Alberty RA. 1955. Physical Chemistry, New York: John Wiley and Sons, p. 157.

Daniels F, Mathews JH, Williams JW, Bender P, and Alberty RA. 1956. Experimental physical chemistry, New York: McGraw-Hill Book Co., pp. 47-511, 370-373.

Enviro Control Inc. 1980. Cost analysis methodology and protocol estimates: environmental standards. Rockville, MD: Enviro Control, Inc., Borriston Laboratories, Inc.

Dilling WL. 1977. Interphase transfer processes. I. Evaporation rates of chloromethanes, ethanes, ethylenes, propanes, and propylenes from dilute aqueous solutions. Comparisons with theoretical predictions. Env. Sci. Tech. 11:405-409.

Edwards CA. 1973. Persistent pesticides in the environment. Cleveland: CRC Press. p. 21.

USEPA. 1975. U.S. Environmental Protection Agency, Office of Pesticide Programs. Proposed guidelines for registering pesticides in the United States. Fed. Regist., June 25, 1975, 40 p. 26889.

USEPA. 1978. U.S. Environmental Protection Agency, Office of Pesticide Programs. Proposed guidelines for registering pesticides in the United States. Fed. Regist., July 10, 1978 43 p. 29711- 29712.

USEPA. 1979. U.S. Environmental Protection Agency, Office of Toxic Substances. Discussion of premanufacture testing policy and technical issues, Fed. Regist. March 16, 1979 44 p. 16240-16292.

Glasstone S. 1946. Textbook of physical chemistry, 2nd ed. New York: D. Van Nostrand Co., p. 446-449.

Hamaker JW and Kerlinger WO. 1969. Vapor pressure of pesticides. Adv. Chem. Series 86:39-54.

Harper LA, White AW, Jr., Bruce RR, Thomas AW, and Leonard RA. 1976. Soil and micro climate effects of trifluralin volatilization. J. Environ. Qual. 5:236-242.

Hartley GS. 1969. Evaporation of pesticides. Adv. Chem. Series 86:115-134.

Heller SR, McGuire JM and Budde WL. 1975. Trace organics by GC/MS. Environ. Sci. Tech. 9:210-213.

International Union of Pure and Applied Chemistry. IUPAC, 1972. Commission on Thermodynamics and Thermochemistry. 1972. A guide to procedures for the publication of thermodynamic data. Pure and Appl. Chem. 29:397-407.

Lichtenstein EP. 1971. Environmental factors affecting fate of pesticides in Degradation of Synthetic Organic Molecules on the Biosphere. Washington, DC: National Academy of Sciences, p.192.

MacKay D and Leinonen PJ. 1975, Rate of evaporation of low-solubility contaminants from water bodies to atmosphere. Environ. Sci. Technol. 9:1178-1180.

MacKay D and Wolkoff AW. 1973. Rate of evaporation of low-solubility contaminants from water bodies to atmosphere. Environ. Sci. Technol. 7:611-614.

Seiber JM, Shafik TM and Enoa HF. 1975. Determination of pesticides and their transformation products in water. In Haque R and Fried VH eds. Environmental dynamics of pesticides, New York: Plenum Press, p.18.

Spencer WF and Cliath MM. 1969. Vapor density of dieldrin J. Agric. Food Chem. 3:664-670.

Spencer WF and Cliath MM. 1970. Vapor density and apparent vapor pressure of lindane. J. Agric. Food Chem. 18:529-530.

Thomson GW and Douslin DR. 1971. Vapor pressure. In: Physical methods of chemistry, Vol. I. Part V., Weissberger A and Rossiter BW, eds. New York: Wiley-Interscience, p. 46-89.

OCTANOL/WATER PARTITION COEFFICIENT

STANDARD FOR TEST DATA DEVELOPMENT
PROPOSED RULE, SECTION 4, TSCA

Refers to

Part 772 — Standards for Development of Test Data
Subpart L. Physical, Chemical and Environmental
Persistance Characteristics
Section 772.122-4 Octanol/Water Partition Coefficient

October 1980

TABLE OF CONTENTS

PAGE

1. NEED FOR THE TEST..... 1

2. SCIENTIFIC ASPECTS..... 3

2.1. Rationale for the Use of the Octanol/Water Partition Coefficient to Estimate Bioconcentration Potential..... 3

2.2. Rationale for the Selection of the Test Method..... 4

(2.2.1.) The Conventional Method of Determining the Octanol/Water Partition Coefficient K_{ow}..... 4

(2.2.2.) Other Experimental Methods of Determining K_{ow}.... 5

(1) Reverse-Phase High-Pressure Liquid Chromatography as a Method of estimating K_{ow}.... 5

(2) Thin-Layer Chromatography as a Method of Estimating K_{ow}..... 7

(3) Estimation of K_{ow} from Water Solubility Data..... 7

2.3. Rationale for the Selection of the Test Conditions..... 9

(2.3.1.) Theory of the Distribution Law and the Octanol/Water Partition Coefficient..... 9

(2.3.2.) Factors which Affect the Value of K_{ow}..... 12

(1) Effect of Temperature..... 12

(2) Purity of the Solvents..... 13

(3) Concentration of Solute..... 13

(4) Equilibration Time..... 14

(5) Octanol/Water Volume Ratio..... 14

(6) Chemical Analysis of the Octanol and Water Phases..... 17

(7) Emulsification and Ultracentrifugation..... 18

(8) Equilibration Vessel..... 18

(9) Speciation Effects..... 19

(10) Presaturation of the Solvents..... 20

PAGE

2.4. Reference Compounds..... 20

2.5. Test Data Required..... 21

2.6. Statistical Analysis of the Data..... 23

3. ECONOMIC ASPECTS..... 25

4. REPLIES TO THE COMMENTS ON THE PUBLISHED SECTION 5 TESTING GUIDELINES..... 27

5. TSCA/FIFRA COMPARABILITY..... 32

6. TSCA/OECD COMPARABILITY..... 33

7. REFERENCES..... 35

1. NEED FOR THE TEST

Bioconcentration, the accumulation of a substance in living tissues or other organic matter as a result of net chemical uptake from the medium (e.g., water), is a factor in determining the movement of a chemical in the environment and the potential effects of the chemical on biota. Hydrophobic chemicals which are present in the aqueous environment at subtoxic concentrations may accumulate to toxic levels once inside organisms, presumably through diffusion into nonpolar cell components where they accumulate because of their greater solubility. Further movement of the substance in living tissues may occur as a result of ingestion of lower trophic level organisms, i.e., food chain effects.

The octanol/water partition coefficient K_{ow} has been shown to be a good predictor of the tendency of chemicals to bioconcentrate in fish (Neely et al. 1974). Since 1974, K_{ow} has been used as a measure of bioconcentration potential in fatty tissues in aquatic and other living organisms. The numerical value of the octanol/water partition coefficient is one factor to be considered in determining whether to conduct fish bioconcentration studies. Other factors must also be taken into account. For example, transformation rates (e.g., rates of biodegradation, hydrolysis, photolysis, and oxidation) must also be considered. If a chemical transforms readily by one of these

processes, the potential for bioconcentration will be reduced significantly and fish bioconcentration studies may not be needed.

The octanol/water partition coefficient has been introduced by Hansch to correlate biological activity and chemical structure (Hansch 1969; Hansch and Fujita 1964). Numerous papers have been published by Hansch and his coworkers on this subject in the ensuing years. A monograph has been published on the Hansch approach (Gould 1972).

A recent publication has indicated that the sorption of several hydrophobic pollutants on natural sediments can be related to the octanol/water partition coefficient. Karickhoff et al. (1979) showed that a reasonable estimate (within a factor of two) of the sorption behavior of hydrophobic pollutants can be made from knowledge of the particle size distribution and associated organic content of the sediment and the octanol/water partition coefficient.

A recent publication has described a novel method for estimating the distribution of a chemical in the environment (Mackay 1979). K_{ow} is used in this partitioning analysis. This partitioning analysis will be used as a guide to ecological and health effects testing.

2. Scientific Aspects

2.1. Rationale for the Use of the Octanol/Water Partition Coefficient to Estimate Bioconcentration Potential

Intuitively, the absorption and fat storage of xenobiotic chemicals in living organisms seem to be related to lipophilicity or preferential solubility in fats as compared to water. By definition, the octanol/water partition coefficient K_{ow} expresses the equilibrium concentration ratio of an organic chemical partitioned between octanol and water in dilute solution. If one assumes that octanol simulates fats in its solubilizing effect on organic chemicals, then K_{ow} should be a potential measure of the ease of storage of organic chemicals in fats. For example, a large value of K_{ow} indicates that an organic chemical is not very soluble in water but soluble in octanol. Hence, this would suggest the potential for a large storage of the organic chemical in fats. Davies et. al. (1975) reported human pesticide poisoning by a fat-soluble organophosphate, dichlofenthion. The octanol/water partition coefficient K_{ow} was found to be very high (1.37×10^5) which correlated with the high fat storage of this chemical.

Neely et al. (1974) found a pronounced correlation between K_{ow} and the bioconcentration in trout muscle. Specifically, these researchers obtained a linear correlation between the log

of bioconcentration and the log of calculated K_{ow} for a series of organic chemicals. Since that time, K_{ow} has been used by researchers as an index of bioconcentration potential in living organisms. The Office of Pesticide Programs (OPP) [EPA 1975, 1978] has proposed, and the Organization for Economic Cooperation and Development (OECD 1978) is considering the use of K_{ow} as a measure of bioconcentration potential in aquatic organisms.

2.2. Rationale for the Selection of the Test Method

2.2.1. The Conventional Method of Determining the Octanol/Water Partition Coefficient K_{ow}

The conventional method for determining a distribution coefficient is carried out by distributing a chemical between two immiscible liquids in a vessel and measuring the concentration of the chemical in the two liquid phases after equilibration (Glasstone 1946; Leo et al. 1971). This method can be applied to the determination of the octanol/water partition coefficient K_{ow}. Numerous researchers use the conventional method of determining K_{ow} and have published papers using this method (e.g., Fujita et al. 1964; Hansch and Anderson 1967; Leo et al. 1971; Chiou et al. 1977). OPP (EPA 1975, 1978) has proposed, and OECD (1978) is considering the conventional method of determining K_{ow}. Most chemical companies that determine octanol/water partitioning use the conventional method of determining K_{ow}.

Hence, the proposed test standard utilizes the conventional method of determining the octanol/water partition coefficient K_{ow}. It should be noted that there is no validated standard test method for determining K_{ow} (e.g., an ASTM method). The proposed method in this standard was developed from a thorough review of the research literature on the experimental determination of K_{ow} and by talking to researchers who have considerable experience in carrying out these experiments.

2.2.2. Other Experimental Methods of Determining K_{ow}

(1) Reverse-Phase High-Pressure Liquid Chromatography as a Method of Estimating K_{ow}. A rapid method based on reverse-phase high-pressure liquid chromatography has been developed by Veith (Veith and Morris 1978; Veith et al. 1979) to estimate the octanol/water partition coefficient of organic chemicals. Using the solvent mixture water/methanol (15/85 v/v) as the elutant, the log of the retention time [log (t_R)] of organic chemicals on a permanently bonded (C-18) reverse-phase high-pressure liquid chromatographic system has been found to be linearly related to log K_{ow}. This relationship has been expressed by the equation

$$\log K_{ow} = A \log(t_R) - B, \qquad (1)$$

where A and B are constants determined from the experimental data for some organic chemicals. Using a mixture of the chemicals benzene, bromobenzene, biphenyl, p,p'-DDE [2,2-bis(p-

chlorophenyl)-1,1-dichloroethylene] and 2,4,5,2',5'-pentachlorobiphenyl, A and B were found to be 5.106 and 1.258, respectively, with a correlation coefficient of 0.975. It must be emphasized that this correlation is limited with respect to being representative of the organic chemicals encountered. This calibration mixture was selected largely on the basis of the log K_{ow} values reported in the literature, and the correlation is linear over five orders of magnitude of K_{ow}. To determine the accuracy of this method of estimating log K_{ow} by comparison with data reported in the literature, Veith and coworkers measured the retention time of 38 chemicals, and the standards and log K_{ow} values were calculated from the regression equation (1). The results indicated that log K_{ow} can be estimated to within (22.8 ± 20.0) percent when compared with the values reported in the literature from measurements using other methods. The percent error was calculated assuming the literature value is the correct log K_{ow}, an assumption these researchers had some reservations about. It should be noted that some of the greatest relative errors were observed with polar chemicals which dissociate in water (e.g., m-chlorobenzoic acid, 2,4,5-trichlorophenol, and diphenylamine). This method has a definite advantage since the estimation of K_{ow} can be made rapidly and relatively easily in comparison to the determination of K_{ow} by the conventional method. Futhermore, K_{ow} can be estimated for individual chemicals in complex mixtures (e.g., solid wastes) without knowing the specific chemical structure of each chemical. Further work is being carried out by Veith (at the Duluth EPA

Laboratory), and others, to develop this method so that it may be applicable to a large number of organic chemicals with a wide range of different organic structures. If this research is successful, this high-pressure liquid chromatographic method of estimating K_{ow} will be proposed as a test standard.

Other researchers have developed high-pressure liquid chromatographic methods to determine K_{ow} (Mirrless et al. 1976; Yamana et al. 1977; Carlson et al. 1975; Hulshoff and Perrin 1976; McCall 1975). However, these methods are based on a very limited number of experiments and considerably more work is needed to develop them.

(2) Thin-Layer Chromatography as a Method of Estimating K_{ow}. It has been reported that thin-layer chromatography can be used to estimate K_{ow} (Mirrless et al., 1976; Hulshoff and Perrin 1976). However, high-pressure liquid chromatography (HPLC) is far superior to thin-layer chromatography (TLC) because of its accuracy (i.e., definition of the peak), reproducibility, ease of detection in many cases, and above all the range of applicability (HPLC is applicable over 5 order of magnitude of K_{ow} while TLC is only applicable over 1.5 orders of magnitude of K_{ow}) (Mirrless et al. 1976).

(3) Estimation of K_{ow} from Water Solubility Data. The octanol/water partition coefficient is defined as the ratio of the equilibrium molar concentation of the chemical in octanol and water. Thus, low molecular mass (i.e., molecular weight)

organic chemicals with a low water solubility should have a high value of K_{ow} (e.g., hydrophobic organic chemicals). Therefore, there should be a correlation between K_{ow} and water solubility. Chiou et al. (1977) studied the relationship between K_{ow} and the water solubility, S, and found that for 34 organic chemicals, an excellent linear correlation was observed between log K_{ow} and log S which extended to more than eight orders of magnitude in water solubility (10^{-3} to 10^{4} ppm), and six orders of magnitude in K_{ow} (10 to 10^{7}). Chiou et al. (1977) found the following regression equation

$$\log K_{ow} = 5.00 - 0.670 \log S, \qquad (2)$$

where K_{ow} is the octanol/water partition coefficient, S is the water solubility in µmol/L, and the correlation coefficient (r^2) was 0.970 for these 34 chemicals. Thus, K_{ow} can be estimated from the experimental value of the water solubility of an organic chemical. This method would have a definite advantage in that K_{ow} could be estimated directly from water solubility data without having to experimentally measure K_{ow}. Thus, the octanol/water test standard could eventually be eliminated, thereby reducing the cost of testing. However, considerably more experimental work is necessary to extend the correlation to a large number of organic chemicals with different structures before it can be used as a test standard. EPA will sponsor research work to develop this method.

2.3. Rationale for the Selection of Test Conditions

A detailed study of the theory of the distribution law, the partition coefficient, and the published literature on the conventional determination of K_{ow} indicates that it is extremely important that numerous factors (or test conditions) be standardized. In order to establish these factors clearly, the theory of the distribution law and its relation to these factors are discussed in detail in the following sections.

2.3.1. Theory of the Distribution Law and the Octanol/Water Partition Coefficient

The distribution coefficient or partition coefficient can be derived using thermodynamic theory (Glasstone 1946). Consider a mixture of two immiscible liquids which is shaken with a solute (organic chemical). The solute distributes itself between the two liquids in such a way that at equilibrium, in dilute solution, the ratio of the concentrations of the solute in the two layers is a constant at a given temperature. The tendency of a chemical to distribute itself between two immiscible liquids with a constant concentration ratio, in dilute solution, is a direct consequence of the thermodynamic requirements for equilibrium. To illustrate this, consider a pair of immiscible liquids A and B in contact with each other containing the same solute in solution. The chemical potential of a solute in solvent A is given by

best copy available

$$\mu_A = \mu^{\circ}_A + RT \ln a_A , \qquad (3)$$

where μ_A is the chemical potential of the solute in solvent A, μ°_A is the standard chemical potential of the solute in the same solvent (i.e., the value of μ_A at $a_A = 1$), while a_A, the activity of the solute in the solvent A is the effective concentration taking into account intermolecular interactions of the solute in the solvent. R is the gas constant and is equal to 8.314 joules/°K/mol, while T is the absolute temperature in °K. Similarly, for solvent B

$$\mu_B = \mu^{\circ}_B + RT \ln a_B, \qquad (4)$$

where all the quantities have the same significance as in equation (3). At equilibrium between the layers $\Delta\mu = 0$; hence

$$\Delta\mu = \mu_B - \mu_A = 0,$$

and

$$\mu_B = \mu_A. \qquad (5)$$

Using equations (3) and (4) in (5) yields

$$\mu^{\circ}_B + RT \ln a_B = \mu^{\circ}_A + RT \ln a_A \qquad (6)$$

$$\ln \frac{a_B}{a_A} = \frac{\mu^{\circ}_A - \mu^{\circ}_B}{RT}.$$

However, at a given temperature, u°_B and u°_A are constants for a given solute in a particular solvent; hence

$$\ln \frac{a_B}{a_A} = K,$$

and,

$$\frac{a_B}{a_A} = K. \qquad (7)$$

Equation (7) is the mathematical statement of the distribution law which states that a substance will distribute itself between two solvents until at equilibrium and the ratio of the activities of a chemical in the two layers is a constant at a fixed temperature, irrespective of the absolute values of a_A or a_B. The activity a_A can be written as

$$a_A = \gamma_A C_A , \qquad (8)$$

where γ_A is the activity coefficient and takes into account the interaction between molecules A in solution, and C_A is the molar concentration. In dilute solution as

$$C_A \rightarrow 0 ,$$
$$\gamma_A \rightarrow 1 ; \qquad (9)$$

hence,

$$\lim_{C_A \rightarrow 0} (a_A) = C_A. \qquad (10)$$

The same argument follows for the solute in solvent B and $\lim_{C_B \rightarrow 0} (a_B) = C_B$. Using these results in equation (7), the distribution coefficient K, in dilute solution, becomes

$$K^{\circ} = \lim_{C \rightarrow 0} \left(\frac{C_B}{C_A} \right) . \qquad (11)$$

For the specific case for the octanol/water partition coefficient, B is the solvent n-octanol, A is the solvent water, and $K^{\circ} = K_{ow}$. Thus, equation (11) becomes

$$K_{ow} = \frac{C_{octanol}}{C_{water}} . \qquad (12)$$

According to Nernst, the distribution law applies only to individual molecular species in solution. If a molecule dissociates or associates in octanol and water, then equation (12) must be modified. In general, if α represents the fraction of the total solute that is dissociated or associated, assuming that either association or dissociation occurs in each solvent, then

$$K_{ow} = \frac{(1 - \alpha_{oct.})\, C_{oct.}}{(1 - \alpha_{water})\, C_{water}} , \qquad (13)$$

since (1-α) gives the fraction of unchanged molecules in each phase. For the special case where no association takes place in octanol, equation (13) reduces to

$$K_{ow} = \frac{C_{oct.}}{(1 - \alpha_{water})\, C_{water}} . \qquad (14)$$

2.3.2. Factors Which Affect the Value of K_{ow}

(1) Effect of Temperature. From the theory of the distribution law as outlined in section 2.3.1., the distribution coefficient K is a function of the temperature (equation (6)), and is a constant at a fixed temperature (equation (7)). Since K_{ow} is a distribution coefficient, it should also vary with temperature and is a constant at a fixed temperature. Hence, in carrying out octanol/water partition coefficient experiments by

the conventional method, the temperature should be controlled. However, variations due to temperature are small compared to those inherent in the errors in the other measurements, e.g., the errors in measuring the concentration of solute in octanol and water. Therefore, for reasonably accurate determinations of K_{ow}, it is sufficient to control the temperature to ± 1°C. Since most physical properties of chemicals are reported at 25°C, the proposed standard requires that K_{ow} be determined at this temperature.

(2) Purity of the Solvents. Trace amounts of impurities present in n-octanol tend to produce emulsions and must be removed (Fujita et al. 1964; Hansch and Anderson 1967; Chiou et al. 1977). Emulsions give poor phase separation and result in a wide scatter in the value of K_{ow}. In addition, impurities in octanol may affect the analysis for the solute. Hence, the proposed test standard required that the octanol be $\geq$99.9% pure. Distilled or reagent grade water (ASTM Type II) should be used.

(3) Concentration of Solute. From the theory of the distribution law, as outlined in section 2.3.1., equations (12), (13), and (14) only apply in dilute solution. Hence the proposed test standard requires that all experiments be carried out at molar concentration C < 0.01M in octanol and water.

(4) Equilibration Time. For many chemicals, 5 minutes of gentle agitation of the two phase system established equilibrium and produced consistent results (Leo et al. 1971). Studies by Craig and Craig (1950) indicated that when the phases were of about equal volume, equilibrium was rapidly attained. When high ratios of water to octanol (>100:1) were used, longer shaking was necessary to establish equilibrium. High ratios of water to octanol are used to determine K_{OW} for very hydrophobic organic chemicals (sections 2.3.2.(5) and (8)) as described below. Therefore, for most chemicals, gentle agitation for 1 hour should be adequate to reach equilibrium. For surfactants, at least 16 hours of agitaton is necessary to reach equilibrium. This is an empirical observation obtained by researchers who have carried out experiments with surfactants. It is undoubtedly due to the nature of surfactant chemicals.

(5) Octanol/Water Volume Ratio. Depending upon the solubility of the solute in octanol and water, the ratio of the volume of octanol to water should be adjusted. For hydrophobic solutes, which are very insoluble in water, considerably more water than octanol should be used. Adjustment of solvent volumes can decrease the effect of analytical errors and consequently decrease the error in determining K_{OW} (Leo et.al. 1971). The following example will illustrate this point. Consider a chemical with a molecular mass (i.e., molecular weight) MW (mg/mmol), K_{OW} = 200. Twenty mg of chemical are dissolved in 100 mL of octanol, and 100 mL of water are added to this system.

After equilibration, the mass of chemical in each phase can be calculated as follows. The chemical will partition with x mg in the water phase and (20-x) mg in the octanol phase.

$$C_{oct.} = \frac{\frac{(20-x)\,mg}{MW\ (mg/mmol)}}{100\ mL} = \frac{(20-x)}{100\ MW}\ \frac{mmol}{mL} = \frac{(20-x)}{100\ MW}\ \text{Molar}$$

$$C_{water} = \frac{\frac{x\ mg}{MW(mg/mmol)}}{100\ mL} = \frac{x}{100\ MW}\ \frac{mmol}{mL} = \frac{x}{100\ MW}\ \text{Molar}$$

Since

$$K_{ow} = \frac{C_{oct.}}{C_{water}},$$

$$200 = \frac{\frac{(20-x)}{100\ MW}}{\frac{x}{100\ MW}} = \frac{(20-x)}{x}.$$

Then $x = \frac{20}{201} = 0.0996 = 0.10$ mg = mass of chemical in the water phase; and $(20-x) = 19.9 \approx 20$ mg = mass of chemical in the octanol phase.

Consider an analytical error of $\pm$ 0.05 mg in the aqueous phase (i.e., 0.10 - 0.05 = 0.05 and 0.10 + 0.05 = 0.15).

$$K_{ow} = \frac{\frac{20}{MW}}{\frac{0.05}{100}} = \frac{20}{0.05} = 400$$

$$K_{ow} = \frac{\frac{20}{MW}}{\frac{0.15}{100}} = \frac{20}{0.15} = 133.$$

Therefore, an analytical error of ± 0.05 mg in the aqueous phase means that K_{ow} can range from 400 to 133, and a very large error in K_{ow} occurs.

Now consider the same example as above but now the solvents are adjusted to 200 mL of water and 5 mL of octanol. After equilbration, the mass of chemical in each phase is now:

$$C_{oct.} = \frac{\frac{(20-x)}{MW}}{5} = \frac{(20-x)}{5\ MW} \text{ Molar}$$

$$C_{H_2O} = \frac{\frac{x}{MW}}{200} = \frac{x}{200\ MW} \text{ Molar}$$

$$K_{ow} = \frac{C_{oct.}}{C_{water}}$$

$$200 = \frac{\frac{(20-x)}{5\ MW}}{\frac{x}{200\ MW}} = \frac{(20-x)}{x} \quad (40)$$

Then $x = \frac{20}{6} = 3.33$ mg = mass of chemical in the water phase; and

(20-x) = 16.7 mg = mass of chemical in the octanol phase.

Now consider the same analytical error of $\pm$0.05 mg in the aqueous phase (i.e., 3.33 - 0.05 = 3.28 and 3.33 + 0.05 = 3.38)

$$K_{ow} = \frac{\frac{16.7}{MW}}{\frac{3.28}{MW}} = \frac{16.7}{3.38} \quad (40) = 203$$

$$K_{ow} = \frac{\frac{16.7}{MW}}{\frac{3.28}{MW}} = \frac{16.7}{3.28} \quad (40) = 197.$$

Now the analytical error of ± 0.05 mg in the aqueous phase means that K_{ow} can range from 197 to 203 and the error in K_{ow} has been reduced dramatically.

(6) Chemical Analysis of the Octanol and Water Phases. Consider a partitioning experiment in which a chemical is dissolved in octanol at a low concentration (less than 0.01 molar). The conventional partitioning experiment is carried out and only one phase is analyzed for the molar concentration of the solute. Using a mass balance, the molar concentration of the solute in the other phase is obtained by difference. However, if there is a loss of chemical by adsorption to the surface of the glass walls, a serious error will occur at this low concentration. This is especially true for very hydrophobic chemicals (Chiou et al. 1977) and for ionic solutes (Leo et al. 1971). Therefore, the proposed test standard requires that both the octanol and water phases be analyzed.

An analytical method should be selected that is the most applicable to the analysis of the specific chemical. However, large errors can occur as a result of traces of more-water-soluble contaminants that are not analytically distinguishable from the parent chemical. This error is very significant when the analytical method is ultraviolet absorption spectroscopy or radiometry, since these methods can be nonspecific for many solutes. Therefore, chromatographic methods are preferable because of their compound specificity in analyzing the parent chemical without interference from

impurities (Karickhoff and Brown 1979). Whenever practicable, the chosen analytical method should have a precision within ± 5 percent.

(7) Emulsification and Ultracentrifugation. Many chemicals can cause troublesome emulsions to form between octanol and water and emulsification can result in large errors in K_{ow} (Leo et al. 1971; Chiou et al. 1977). This is especially true for hydrophobic chemicals. Therefore, gentle shaking shall be used to minimize the formation of emulsions. In addition, incomplete separation of the two phases is one of the most serious sources of error. To break any emulsion formed and to completely separate the octanol and water phases, the proposed test standard requires that the two phase system be ultracentrifuged at 25°C for 20 minutes. The acceleration G value required to break an emulsion and to completely separate the octanol and water phases must be determined by trial-and-error experimentation. Since the clarity of the two phases is not a dependable criterion of the absence of an emulsion and complete separation of the two-phase system, the proposed test standard requires that a turbidimeter be used to make sure that the emulsion is broken and the octanol and water phases have been completely separated.

(8) Equilibration Vessel. To simplify the experimental procedure, equilibration should be carried out in a centrifuge tube (special glass tubes can be used up to approximately 12,000 G and stainless steel centrifuge tubes can be used at higher G values) with a sealable cap. This will avoid a transfer step and volatile

chemicals can be handled easily. The centrifuge tube shall be almost completely filled with the two phase mixture to minimize partitioning with air. This is especially important when determining K_{ow} for volatile chemicals (Hansch and Anderson 1967).

Very hydrophobic chemicals, with K_{ow} in the order of 10^4 to 10^6, require relatively large volumes of the aqueous phase (section 2.3.2.(5)). Hence, for these chemicals, the proposed test standard requires that equilibration be carried out in a large ground-glass stoppered flask.

(9) Speciation Effects. The details of speciation have been discussed in the theory of the distribution law and the octanol/water partition coefficient, section 2.3.1.

If the chemical does not associate or dissociate in octanol and water, then the proposed test standard requires that equation (12) be used and K_{ow} be determined at concentrations $C < 0.01M$ and $C_1 = 0.1C$. Under these experimental conditions, if K_{ow} is constant, then association or dissociation has been minimized or eliminated.

If the chemical associates in octanol or water or in both liquids, then the proposed test standard requires that equation (13) be used and K_{ow} be determined at concentations $C < 0.01M$, $C_1 = 0.1C$, $C_2 = 0.01C$, $C_3 = 0.001C$, ... When K_{ow} is constant at two concentrations differing by a factor of 10, then the effect of association has been minimized or eliminated.

For chemicals which reversibly ionize or protonate (e.g., carboxylic acids, phenols or anilines), the proposed test standard requires that equation (14) be used with water buffered at pH 5.0, 7.0, and 9.0, the pHs of environmental concern.

(10) Presaturation of the Solvents. Presaturation of octanol with water and water with octanol is required for this test standard. The preparation of these saturated solutions is very simple to carry out. This requirement is extremely important when determining K_{ow} for very hydrophobic chemicals since the ratio of water to octanol will be very large. In this case, if the experiment is carried out without presaturation of the water with octanol, then all the octanol will dissolve in the aqueous phase and K_{ow} cannot be determined.

2.4. Reference Compounds

It would be very desirable to have reference compounds which cover a K_{ow} range of 10 to 10^6. These reference compounds would provide the experimenter with comparative reference values to determine how well the test has been conducted. Unfortunately, these reference compounds are not available. The EPA is funding work at the National Bureau of Standards to develop such reference compounds. When this work is completed, these reference compounds will be recommended for use in this test standard. In the interim, it is recommended that the book by Hansch and Leo (1979) be used for the selection of potential reference compounds.

2.5. Test Data Required

The tendency of an organic chemical to partition out of water into other environmental compartments containing hydrophobic constituents (e.g., aquatic organisms) can be inferred from the values of the octanol/water partition coefficient K_{ow}. Chiou et al. (1977) developed regression equations relating log K_{ow} with water solubility S (in μmol/L) and bioconcentration in rainbow trout (BF). Assuming log K_{ow} is between 1 and 6, S and BF can be calculated; these results are summarized in Table 1 (note that S has been converted to mol/L). Furthermore, assuming that the average molecular mass (i.e. molecular weight) of an organic chemical is 300 gm/mol, the water solubility can be converted to ppm; these results are also summarized in Table 1. It is apparent that for log K_{ow} = 6 (i.e., $K_{ow} = 10^6$), the water solubility will be extremely low (9.7×10^{-3} ppm or 9.7 ppb) and the predicted BF is 1.48×10^4. Hence, the data indicate that the chemical will partition out of the water phase and into the fat of the fish (i.e., the hydrophobic phase). For log K_{ow} = 1 (i.e., K_{ow} = 10), the water solubility will be very high (2.80×10^5 mg/L or 280 gm/L) and the predicted BF is 2.4. Hence, these data indicate that the chemical will remain in the water phase and will not partition significantly into the fat of the fish (i.e., the hydrophobic phase). Therefore, the proposed test standard is designed to determine the value of K_{ow} in the range 10 to 10^6. Low molecular mass organic chemicals with a K_{ow} value less than 10 will not partition significantly into, or tend to

Table 1. Summary of Calculated Values of Water Solubility and Bioconcentration in Rainbow Trout as a Function of Log K_{ow}[a]

log K_{ow}	S(mol/L)[b]	S(mg/L or ppm)[c]	BF[d]
6	3.24×10^{-8}	9.7×10^{-3}	1.48×10^{4}
5	1×10^{-6}	0.30	2.57×10^{3}
4	3.09×10^{-5}	9.3	4.47×10^{2}
3	9.77×10^{-4}	2.93×10^{2}	7.76×10^{1}
2	3.02×10^{-2}	9.06×10^{3}	1.35×10^{1}
1	9.33×10^{-1}	2.80×10^{5}	2.4

a Regression equations taken from Chiou et al. (1977).
b Water solubility
c Water solubility in ppm assuming a molecular mass of an organic chemical is 300 gm/mol.
d Bioconcentration in rainbow trout

accumulate in, any hydrophobic environmental compartments. Low molecular mass organic chemicals with K_{ow} in excess of 10^6 will tend to accumulate into all hydrophobic enviromental compartments. For low molecular mass organic chemicals outside the range 10 to 10^6, the proposed test standard requires that K_{ow} be characterized as < 10 or > 10^6 with no further quantification.

Specific analytical and recovery procedures are needed to determine whether acceptable data have been generated.

2.6. Statistical Analysis of the Data

Numerous researchers have published data on the determination of the octanol/water partition coefficient by the conventional method (e.g., Chiou et al. 1977; Davies et al. 1975; Fujita et al. 1964; Hansch and Anderson 1967; Leo et al. 1971). However, none of these researchers has analyzed the data statistically and the precision of K_{ow} as determined by the conventional method has not been clearly established. The precision is, in part, a function of the nature of the specific chemical. As the hydrophobicity of the chemical increases, K_{ow} increases and the precision of K_{ow} decreases. Furthermore, the precision is also a function of the analytical procedure used. In general, the lower the concentration to be measured, the poorer is the precision of the analytical procedure. Therefore, no reliable precision can be stated at this time for determining K_{ow}. Obviously, the precision can be improved by making numerous

replicate determinations. However, in order to minimize cost, it has been decided to determine K_{ow} with three replicates. The submitter of the test results must analyze the data statistically. When a large number of chemicals have been determined by the proposed method, the data will be analyzed statistically and the level of precision can be defined for various ranges of K_{ow}.

3. ECONOMIC ASPECTS

A survey of three commercial laboratories to estimate costs for performing the test outlined in this Standard found a range of $100 to $1,400 and with a "best estimate" of $553. A cost estimate was also made by separating the protocol into components and estimating the cost of each component including direct labor cost, overhead cost, other direct costs, general and administrative costs, and profit or fee. The estimated range for this test ranged from $229 to $688 with a "best estimate" of $458. The cost estimates were made assuming that all the requirements of Good Laboratory Practice Standards, as specified in section (c) of this Test Standard, are being satisfied. Details of the cost estimates are contained in a report by Enviro Control, Inc. (1980).

The cost estimates in the Enviro Control report do not include the costs for specialized analytical methodology or for analytical method development that is required to determine the concentration of the chemical in octanol and water at equilibrium. It is not possible to estimate the cost of such special analytical methodology since the costs are related to the nature of the chemical to be analyzed and related because such method development costs may be prorated among the analytical needs of other test standards.

In reviewing the state of technology concerning this area of testing, the Agency has not identified any adequately developed

tests that would be significantly less costly while providing comparable information.

4. REPLIES TO THE COMMENTS ON THE PUBLISHED SECTION 5 TESTING GUIDELINES

The Federal Register of March 16, 1979 (EPA 1979) presented a discussion of the policy issues, alternative approaches, and test methods under consideration as guidance for premanufacture testing. Included in the test methods was a section on the octanol/water partition coefficient, K_{ow} (pages 16254-16256), which discussed a method for determining this parameter for use in developing information on new chemicals. Among the comments received in response to the discussion were several comments related to the determination of K_{ow}.

One commentor made the following statement: "The rationale for including this test in the proposed premanufacture testing program is based upon published data indicating that there is a correlation of K_{ow} with bioconcentration or biomagnification potential of chemical substances in aquatic and other living organisms. However, researchers (Chiou et al. 1977; Lu and Metcalf 1975; Metcalf et al. 1975) have shown that there is a correlation between bioconcentration and aqueous solubility suggesting that the assessment for bioconcentration potential can be obtained from water solubility data. Under these circumstances, the determination of K_{ow} would be redundant." However, K_{ow} has other uses in addition to predicting bioconcentration potential. K_{ow} is related to partitioning into all hydrophobic environmental compartments (e.g., sediments). In addition, K_{ow} is an indicator of biological activity. Therefore, K_{ow} is a very

important and useful physical/chemical property. The research work by Chiou is significant because K_{ow} can be estimated from water solubility data and this work has been thoroughly discussed in section 2.2.2. (3). Research work will be sponsored by EPA to develop this method.

Another commentor indicated that HPLC has been overlooked as an alternative method for determining K_{ow}. However, this method has been addressed in the TSCA Federal Register guidelines and has been thoroughly discussed in section 2.2.2. (1). HPLC is much simpler to run than the conventional method. Hence, research will be sponsored by EPA to develop this method.

Two commentors indicated that the range of K_{ow} should be limited. One commentor suggested that the range should be 10 to 10^4 while another commentor indicated that there is very little value in pushing the method to 10^6 or to very low values. This subject has been thoroughly discussed in section 2.5. The data presented in this section indicated that the most useful range of K_{ow} is 10 to 10^6 and for values outside this range, K_{ow} should be characterized by <10 or $>10^6$ without further quantification. The value of K_{ow} proposed by one commentor (i.e., 10^4) is too low since a significant amount of a chemical would still be in the water phase, bioconcentration is moderate (447), and partitioning is not complete. It is only when $K_{ow} = 10^6$ that the water solubility is very low (9.7 ppb), bioconcentration is high (1.48×10^4) and partitioning into a hydrophobic environmental compartment is essentially complete.

There were two comments related to equilibration time. One commentor indicated that the equilibrium time of 1 hour is entirely too short since it can take several weeks or months to establish equilibrium in a water solublity experiment for very hydrophobic chemicals. However, in determining water solubility, the equilibration is between the molecules in the solid state and in solution and this process is very slow for very hydrophobic chemicals. On the other hand, in determining K_{ow}, the chemical is dissolved in octanol, water is added, and equilibrium is involved with molecules in solution. Under these conditions, equilibration is rapid (5-15 minutes). Equilibrium has been discussed in section 2.3.2. (4).

Another commentor indicated that in order to ensure that equilibrium has been established, several measurements of K_{ow} are necessary at different times, e.g., 1, 6, 16 hours. However, this procedure would be too costly since several determinations must be made. As indicated in section 2.3.2. (4), equilibrium is attained rapidly for most chemicals (5-15 minutes) and 1 hour is sufficient to reach equilibrium. For surfactants, 16 hours is sufficient to reach equilibrium.

Three commentors discussed the determination of K_{ow} at pH 5, 7, and 9. One commentor indicated that the measurement of K_{ow} at different pHs is inappropriate since the objective of this measurement is to simulate bioaccumulation in fatty tissues under physiological and surface water conditions (pH 7). This statement is not true. In general, the pH of surface waters can vary

from 5 to 9 and is not necessarily at 7. Hence for chemicals which reversibly ionize or protonate, the pH of the surface water will determine the type of molecular species present and this molecular species is the one which will be bioconcentrated. In addition, this test will give information on sorption into any hydrophobic compartment (e.g., sediments) which is a function of the nature of these molecular species and, therefore, will have a direct effect on partitioning in the environment. Another commentor suggested that it would be more than sufficient to measure the partition coefficient of the chemical at the pH (5 or 9) that will give the lowest solubility since this will most likely yield the highest partition coefficient. However, K_{ow} will be used in a partitioning analysis and it is necessary to know K_{ow} at pH 5, 7, or 9, the pHs of environmental concern. The third commentor suggested that the same buffers described in the hydrolysis protocol (p. 16268) be used. The buffers listed in the hydrolysis procedure were designed to minimize buffer effects. Hence, these buffers have been recommended for use in the proposed test standard.

Two commentors discussed the use of glass-stoppered centrifuge tubes at 37,000 G. They indicated that currently there are no glass-stoppered centrifuge tubes that can be used at 37,000 G. This suggestion has been incorporated in the proposed test standard. Special glass centrifuge tubes are available commercially which can be used to approximately 12,000 G and stainless steel centrifuge tubes can be used at higher G values.

One commentor discussed the inadequacy of the sampling procedure. The commentor indicated that since the octanol layer is on top of the water layer, it would be extremely difficult to sample the aqueous phase without contaminating the sampling device (the pipet) with the concentrated octanol phase. The test standard has incorporated a method which will avoid this problem. The test standard requires that the octanol phase be sampled first with a pipet. Then the remainder of the octanol phase and the interfacial layer can be removed and discarded. Finally, a new pipet can be used to sample the aqueous layer.

One commentor discussed the problem of emulsification. It was indicated that even gentle shaking for 1 hour could lead the same horrendous emulsions for certain chemicals. This problem has been recognized and thus ultracentrifugation is required to break any emulsion and to separate the octanol and water phases.

5. TSCA/FIFRA COMPARABILITY

The methods for determining the octanol/water partition coefficient described in the FIFRA Guidelines (EPA 1975, 1978) and this Test Standard are similar. However, the FIFRA Guidelines only give general guidance for determining K_{ow}. Because the determination of K_{ow} is a difficult procedure, as explained above, a number of variables must be controlled in order to obtain a reliable result. Therefore, detailed procedures for controlling these variables have been included in the TSCA Test Standard. Furthermore, adherence to these detailed procedure is required to insure that a uniform method is used by all submitters, thereby allowing the data to be compared.

6. TSCA/OECD COMPARABILITY

The Organization for Economic Cooperation and Development issued a draft of a test protocol for determining K_{ow} (OECD, 1978). This draft is old but is the only one available for comparison with the TSCA proposed test standard. In general, the OECD test protocol and the TSCA proposed standard are very similar. However, there are a few differences which are discussed below. OECD requires that K_{ow} be determined quantitatively regardless of the value for a specific chemical. The TSCA test standard specifies that K_{ow} only be determined quantitatively in the range 10 to 10^6. The justification for this range is outlined in section 2.5.

OECD indicates that K_{ow} is "ideally" dependent on temperature. K_{ow} is, indeed, a function of temperature as discussed in section 2.3.2. However, OECD indicates that the effect of temperature on K_{ow} is small (i.e., approximately 0.01 logarithmic units per degree celsius and can be positive as well as negative). Hence, OECD does not require temperature control and their test method specifies that K_{ow} be measured at room temperature. However, no experimental data are given to support this finding. Since K_{ow} is an equilibrium constant and is dependent on temperature (section 2.3.2.), the TSCA proposed test standard requires that K_{ow} be determined at 25 $\pm$ 1°C. Precise temperature control is not required as indicated by the range of temperature control of $\pm$ 1°C.

OECD does not require that K_{ow} be determined as a function of pH for chemicals that reversibly ionize or protonate. The TSCA proposed test standard requires that chemicals that reversibly ionize or protonate should be tested at pHs 5, 7, and 9, the pHs of environmental concern. The effect of pH is discussed in section 2.3.2. (1).

OECD does not indicate how to handle volatile chemicals or very hydrophobic chemicals while the TSCA test standard gives detailed procedures for handling these types of chemicals. The reasons for giving precise directions on handling these types of chemicals are discussed in section 2.3.2. (8).

OECD requires an equilibration time of 5 minutes while the TSCA proposed standard requires an equilibrium time of one hour for most chemicals and 16 hours for surfactants. Equilibration time is discussed in section 2.3.2. and also has been discussed in section 4.

OECD indicates that the reproducibility is in general $\pm$ 3 percent. However, no data are given to substantiate this finding. A thorough literature search has been made and no data have been published which have established the precision of K_{ow} for a large number of chemicals. Hence, no precision is specified in the TSCA proposed test standard. The statistical analysis of the data is discussed in section 2.5.

8. REFERENCES

Carlson RM, Carlson RE, Kopperman HL. 1975. Determination of partition coefficients by liquid chromatography. J. Chromatogr. 107:219.

Chiou CT, Freed VH, Schmedding DW, Kohnert RL. 1977. Partition coefficient and bioaccumulation of selected organic chemicals. J. Environ. Sci. Tech. 11:475.

Craig LC , Craig D. 1950. In: Technique of organic chemistry, Vol. III, pt. I, Chapter 4. New York: Interscience Publishers, Inc.

Davies JE, Barquet A, Freed V, Haque R, Morgade C, Sonneborn RE, Vaclavek C. 1975. Human poisonings by a fat-soluble organophosphate insecticide. Arch. Environ. Health 30:608.

Enviro Control, Inc. 1980. Cost analysis methodology and protocol estimates; Environmental Review Division standards. Rockville, Md., Enviro Control, Inc., Borriston Laboratories, Inc.

USEPA. 1975. U.S. Environmental Protection Agency. Office of Pesticide Programs. Proposed guidelines for registering pesticides in the United States. Fed. Regist. 1975 40, 26802.

USEPA. 1978. U.S. Environmental Protection Agency, Office of Pesticide Programs. Proposed guidelines for registering pesticides in the United States. Fed. Regist. 1978 43, 29696.

Fujita T, Iwasa J, Hansch C. 1964. A new substituent constant, derived from partition coefficients. J. Am. Chem. Soc. 86:5175.

Glasstone S. 1946. Textbook of physical chemistry. New York: Van Nostrand Co.

Gould RF ed. 1972. Biological correlations--the Hansch approach. Adv. Chem. Ser. No. 114. Washington, D.C.: American Chemical Society.

Hansch C. 1969. A quantitative approach to biomedical structure-activity relationships. Acc. Chem. Res. 2:232.

Hansch C, Anderson SM. 1967. The effect of intramolecular hydrophobic bonding on partition coefficients. J. Org. Chem. 23:2583.

Hansch C, Fujita T. 1964. ρ-α-π anaysis. A method for the correlation of biological activity and chemical structure. J. Am. Chem. Soc. 86:1616.

Hansch C, Leo A. 1979. Substituent constants for correlation analysis in chemistry and biology. New York: J. Wiley & Sons.

Hulshoff A, Perrin JH. 1976. A comparision of the determination of partition coefficients of 1,4-benzodiazepines by high-performance liquid chromatography and thin-layer chromatography. J. Chromatogr. 129:263.

Karickhoff SW, Brown DS. 1979. Determination of octanol/water distribution coefficients, water solubilities, and sediment/water partition coefficients for hydrophobic organic pollutants. EPA-600/4-79-032.

Karickhoff SW, Brown DS, Scott TA. 1979. Sorption of hydrophobic pollutants on natural sediments. Water Res. 13:241.

Leo A, Hansch C, Elkins D. 1971. Partition coefficients and their uses. Chem. Rev. 71:525.

Lu PY, Metcalf RL. 1975. Environmental fate and biodegradability of benzene derivatives as studied in a model aquatic ecosystem. Environ. Health Perspect. 10:269.

Mackay D. 1979. Finding fugacity feasible. Environ. Sci. Tech. 13:1218.

Metcalf RL, Sanborn JR, Lu PY, Nye D. 1975. Laboratory model ecosystem studies of the degradation and fate of radiolabelled tri-, tetra-, and pentachlorobiphenyl compared with DDE. Arch. Environ. Cont. 3:151.

McCall JM. 1975. Liquid-liquid partition coefficients by high-pressure liquid chromatography. J. Med. Chem. 18:549.

Mirrless MS, Moulton SJ, Murphy CT, Taylor PJ. 1976. Direct measurement of octanol-water partition coefficients by high-pressure liquid chromatography. J. Med. Chem. 19:615.

Neely WB, Branson DR, Blau GE. 1974. Partition coefficient to measure bioconcentration potential of organic chemicals in fish. Environ. Sci. Tech. 8:113.

OECD-Chemicals Testing Program. 1978. Draft test protocol for the determination of the partition coefficient of solid and liquid substances in the system water/n-octanol.

Veith GD, Morris RT. 1978. A rapid metnod for estimating log P for organic chemicals. EPA-600/3-78-049.

Veith GD, Austin NM, Morris RT. 1979. A rapid method for estimating log P for organic chemicals. Water Res. 13:43.

Yamana T, Tsuja A, Miyamoto E, Kubo O. 1977. Novel method for determination of partition coefficients of penicillins and cephalosporins by high-pressure liquid chromatography. J. Pharm. Sci. 66:747.

SOIL THIN LAYER CHROMATOGRAPHY

STANDARD FOR TEST DATA DEVELOPMENT
PROPOSED RULE, SECTION 4, TSCA

Refers to

Part 772 — Standards for Development of Test Data
Subpart L. Physical, Chemical and Environmental
Persistance Characteristics
Section 772.122-5 Soil Thin Layer Chromatography

October 1980

CONTENTS

		Page
1.	NEED FOR THE TEST	1
2.	SCIENTIFIC ASPECTS OF SOIL LEACHING	2
	2.1. Introduction	2
	2.2 Basic Processes Affecting Soil Leaching	2
	2.3 Chemical Properties Affecting Leaching	4
	2.4 Soil Properties Affecting Leaching	4
	2.5 Types of Adsorptive Forces	7
	2.6 Surface Transformations	8
3.	ECONOMIC ASPECTS	11
4.	SCIENTIFIC ASPECTS OF THE TEST	12
	4.1 Development of Soil Thin Layer Chromatography (TLC)	12
	4.2 Rationale for the Selection of Soil TLC	15
	4.3 Rationale for Selection of Experimental Conditions and Procedures	17
	4.4 Replies to Federal Register	20
5.	TSCA/FIFRA/OECD COMPARABILITY	23
6.	REFERENCES	24

1. NEED FOR THE TEST

Leaching of chemicals through soil is an important process which affects a chemical's distribution in the environment. If a chemical is tightly adsorbed to soil particles, it will not leach through the soil profile but will remain on the soil surface. If a chemical is weakly adsorbed, it will leach through the soil profile and may reach ground waters and then surface waters. Knowledge of the leaching potential is essential under certain circumstances for the assessment of the fate of chemicals in the environment.

Chemical leaching also affects the assessment of ecological and human health effects of chemicals. If a chemical reaches ground water, deleterious human health effects may arise due to the contamination of drinking water. If a chemical remains at the soil surface, deleterious environmental and human health effects may arise due to an increased concentration of the chemical in the zone of plant growth, possibly resulting in contamination of human food supplies.

Soil thin layer chromatography (TLC) is a qualitative screening tool suitable for obtaining an estimate of chemical's leaching potential. This test is the first of several tests which will be used in obtaining a rough estimation of a chemical's leaching potential and is the test most amenable to standardization at the present time.

2. SCIENTIFIC ASPECTS OF SOIL LEACHING

2.1. Introduction

Since chemical leaching in soils is affected by a large number of interacting processes, this section of the support document will discuss these processes as they relate to this phenomenon.

2.2. Basic Processes Affecting Soil Leaching

As it occurs in the real world, leaching through soil is a complex phenomenon consisting of several major processes (Hamaker 1975). These processes are operative in the soil TLC method and will be discussed in detail below.

The general equation (Guenzi 1974) for chemical movement through porous media under steady state soil-water flow conditions is:

$$\frac{B\delta S}{\theta\delta t} + \frac{\delta c'}{\delta t} = D' \frac{\delta^2 c'}{\delta x^2} - V \frac{\delta c'}{\delta x} \qquad (1)$$

where B = soil bulk density (g/cm^3)

θ = volumetric water content (cm^3/cm^3)

S = amount of chemical adsorbed at the soil/water interface (g/g soil).

t = time (sec.)

C'= solution concentration of chemical

D'= dispersion coefficient (cm^2 /sec)

V = average pore-water velocity (cm/sec)

X = space coordinate measured normal to the section

Most mass transport equations represent simplifications of "real world" conditions. Equation 1 and similar mathematical expressions try to describe the chromatographic distribution of the chemical in the soil profile and are gross simplifications of a phenomenon affected by a number of complex interacting processes including but not limited to precipitation, evaporation, evapotranspiration and hydrodynamic dispersion.

In general, chemical leaching is dependent upon three major processes: the mass transport of water (the direction and rate of water flow), diffusion, and the adsorption characteristics of the chemical in soil (Guenzi 1974). Diffusion is the transport of matter resulting from random molecular motion caused by molecular thermal energy. This random motion will lead to the uniform distribution of molecules in a closed system since there is net movement from regions of higher to lower concentrations. In this document, adsorption refers to the equilibrium distribution of a molecule between a solid phase and a solution phase. As the degree of adsorption increases the concentration of the chemical in the soil water and the soil air decreases. This equilibrium process is governed by two opposing rate processes. The adsorption rate is the rate to which molecules from the liquid phase transfer into the adsorbed state in the solid phase. The desorption rate is the opposite process, i.e.,

the rate at which molecules transfer from the adsorbed state in the solid phase into the liquid phase. In general, the mass transport, diffusion, and adsorption processes produce the observed leaching pattern of a chemical in soil.

2.3. Chemical Properties Affecting Leaching

The main process of the three processes discussed above which determines a chemical's leaching potential (as described mathematically in equation 1) is adsorption. Adsorption is governed by the properties of both the adsorbent and the adsorbate. The important properties of the absorbate affecting adsorption by soil colloids (Bailey and White 1970) are: (1) chemical structure and conformation (2) acidity or basicity of the molecule (pk_a or pk_b), (3) water solubility, (4) permanent charge, (5) polarity, (6) molecular size, and (7) polarizability. There are many ways in which each of these adsorbate properties interact and are manifested in the overall adsorption reaction (Bailey and White 1970).

2.4. Soil Properties Affecting Leaching

Soil is the unconsolidated organic and mineral material on the immediate surface of the earth which serves as a natural medium for the growth of plants. The combined actions of climate, microorganisms and macroorganisms over long periods of time on different parent geologic and biotic materials form soils that differ widely in their physical, chemical, and morphological

characteristics. The wide variations in the amounts and types of clay and organic matter, soil pH, primary and secondary minerals, structure, texture, and exchange capacity create soils of substantial heterogeneity within the United States. There are currently 10 Soil Orders, at least 43 Suborders, over 200 Great Groups and over 7,000 soil series recognized in the United States (Buckman and Brady 1969). The test rule will specify the minimum number of soils depending on the number of sites of manufacture, distribution, use, and disposal of the test chemical. The soil physical/chemical property ranges for pH, organic matter and cation exchange capacity will be specified so that soils used in the TLC method will be representative of U.S. humid and semiarid region mineral soils.

The soil properties affecting the adsorption and desorption of organics include organic matter content, type and amount of clay, exchange capacity, and surface acidity (Adams 1973; Bailey and White 1970; and Helling 1970). Soil organic matter is a primary soil parameter responsible for the adsorption of many pesticides. Helling (1970) lists many examples where the organic matter primarily influenced the adsorption of pesticides. Although organic matter and clay are the soil components most often implicated in pesticide adsorption, the individual effects of either organic matter or clay are not easily ascertained. Since the organic matter in most soil is intimately bound to the clay as a clay-metal-organic complex (Stevenson 1973), two major types of adsorbing surfaces are normally available to the chemical, namely, clay-organic and clay alone. Clay and organic

matter function more as a unit than as separate entities and the relative contribution of organic and inorganic surfaces to adsorption will depend on the extent to which the clay is coated with organic substances. Comparative studies between known clay minerals and organic soils suggest that most, but not all, pesticides have a greater affinity for organic surfaces than for mineral surfaces (Stevenson 1973). Since typical soil studies compare soils in which both clay and organic matter increase and do not utilize multiple regression analyses to isolate the governing parameter (Helling 1970), only generalizations concerning the relative importance of clay and organic matter can be made.

The activity of protons in the bulk suspension (i.e., as measured by pH) and the activity of protons at or in close proximity to the colloidal surface (i.e., the acidity in the interfacial region) may differ significantly. The term "surface acidity" as applied to soil systems is the acidity at or in close proximity to the colloidal surface and reflects the ability of the system to act as a Lewis acid. Surface acidity is a composite term which reflects both the total number of acidic sites and their relative degree of acidity. Surface acidity is probably the most important property of the soil or colloidal system in determining the extent and nature of adsorption of basic organic chemicals as well as determining if acid-catalyzed chemical transformation occurs (Bailey and White 1970). There is overwhelming evidence, mainly from infrared studies, pointing to the fact that there is protonation of basic chemicals by clays

having hydrogen and aluminum as the predominant exchangeable cation and by clays saturated with alkali, alkaline earth, and transition metal cations. A summary of recent investigations indicates that the protonation of chemicals in the interfacial region of clays is a function of the basicity of the molecule, the nature of the exchangeable cation on the clay, water content of the clay system, and the origin of negative charge in the aluminosilicate clay (Bailey and White 1970).

In summary, the chemical properties discussed in (c) and the soil properties discussed in (d) both govern the extent of adsorption in soils.

2.5 Types of Adsorptive Forces

The specific type of interaction of organic molecules with soil will depend on the specific chemical properties of the organic molecule and the type of soil. These specific interactions or adsorptive forces are usually classified as: van der Waals forces, charge transfer, ion exchange, and hydrophobic bonding (Adams 1975, Goring and Hamaker 1972).

The van der Waals forces arise from the fluctuations in a molecule's electron distribution as the electrons circulate in their orbitals. These fluctuations produce instantaneous dipoles which cause that molecule's attraction to other atoms and molecules. Charge transfer involves the formation of a donor-acceptor complex between an electron donor molecule and an electron acceptor molecule with partial overlap of their

respective molecular orbitals and a partial exchange of electron density. Hydrogen bonding is an example of a particular type of charge transfer. Ion exchange refers to the exchange between counterions balancing the surface charge on the soil colloid and the ions in the soil solution. The driving force for this interaction is the requirement for electroneutrality: the surface electric charge must be balanced by an equal quantity of oppositely charged counterions. In general, ion exchange is reversible, diffusion controlled, stoichiometric and, in most cases, exhibits some selectivity or preferential adsorption for one ion over another competing ion. Hydrophobic bonding refers to the preference of an organic molecule for a hydrocarbon solvent or hydrophobic region of a colloid over a hydrophilic solvent. This preference is due to the fact that hydrocarbon regions of a molecule have greater solubility in liquid hydrocarbons (or most organic solvents) than in water. In general, one or more of these specific interactions or adsorptive forces may occur at the same time depending on the presence and magnitude of the chemical and soil properties discussed above.

2.6. Surface Transformations

A special type of interaction between organic molecules and soils deals with the transformation of organic chemicals into new compounds containing different chemical structures through the catalytic activity of the soil colloid surfaces. Although several theories exist to account for the mechanism of these transformations, no scheme predicting the occurrence of such

surface reactions presently exists. Therefore, parent compound mass balances shall be performed and reported as part of the requirements of this soil TLC Test Standard in order to ascertain the extent of such transformations during soil leaching experiments. Also, the leaching pattern (a diagram or photograph of the TLC plate showing the position of the chemical) can give a qualitative indication of the extent of such transformations and shall be reported. The scientific literature shows that a number of chemicals and chemical classes undergo colloid surface induced chemical transformations. Poly-(dimethylsiloxane) fluids in intimate contact with many soils undergo siloxane bond redistribution and hydrolysis, resulting in the formation of low molecular weight cyclic and linear oligomers (Buch and Ingebrightson 1979). S-triazines (White 1976) and organophosphorus pesticides (Yaron 1978, and Mingelgrin et al. 1977) undergo clay colloid induced hydrolysis. Benzene and phenol polymerize into high molecular weight species by adsorption and reaction at the surface of smectite saturated with transition metal cations (Mortland and Halloran 1976). Gallic acid, pyrogallol, protocatechuic acid, caffeic acid, orcinol, ferulic acid, p-coumaric acid, syringic acid, vanillic acid and p-hydroxybenzoic acid undergo oxidative polymerization in the presence of various clay minerals (Wang and Li 1977 and Wang et al. 1978). In general, testing methods that do not take into account surface

transformations should not be used in determining the leaching potential of chemicals.

In summary, the interfacial region is important in determining the adsorption mechanism, the energy by which the adsorbate is held, and in determining if the adsorbed chemical is transformed. This information is important in determining the persistence and ultimate toxicity of the molecule since the transformation product(s) (1) may be more or less toxic than the original compound, (2) may be more or less tightly bound than the original compound, and (3) may have a water solubility either greater than or less than the original compound, thereby affecting its leaching and movement into the groundwater.

3. ECONOMIC ASPECTS

A survey of commercial laboratories to estimate costs for performing the test outlined in this standard found a range of $390-$850 with a "best estimate" of $620. A cost estimate also was made by separating the standard into components and estimating the cost of each component including direct labor cost, overhead cost, other direct costs general and administrative costs and profit or fee. The best estimate of cost was $344 with an estimated range of $172 to $516. An analysis of the discrepancy between the price estimate and survey pointed out that the nature of the method of analysis, after solvent migration, has an impact on the cost of analysis. Different methods will cause variations in cost. Also, the price estimate does not include costs for any analytical work which may be necessary before the test can be performed. In practice, testing laboratories receive chemicals in a pure form, and most identification and analytical methodology has already been developed. Extraction, GC, MS, etc. methodology may be necessary, however, and it is estimated that this would cost less than $1000 for 90 percent of the chemicals tested.

The above cost estimates were made assuming that all the requirements of Good Laboratory Practice Standards (GLPs) are being satisfied. Details of the cost estimate are contained in a report by Enviro Control, Inc. (1980).

4. SCIENTIFIC ASPECTS OF THE TEST

4.1. Development of Soil Thin Layer Chromatography (TLC)

Before 1968, methods of investigating the mobility of nonvolatile organic chemicals within soils were based on the use of field analysis, soil adsorption isotherms, and soil columns. In 1968, Helling and Turner introduced soil thin layer chromatography (soil TLC) as an alternate procedure. It is analogous to conventional TLC, with the use of soil instead of silica gels, oxides, etc. as the adsorbent phase.

In their initial report, Helling and Turner used Lakeland sandy loam, Chillum silt loam, and Hagerstown silty clay loam. Medium sand (>250 µm dia.) was removed from Chillum and Hagerstown soils and coarse sand (>500 µm) from Lakeland soil by dry-sieving. Aqueous slurries were prepared and 500 um (silt loam, silty clay loam) or 750 µm (sandy loam) thick layers were spread on TLC plates using conventional TLC apparatus. After drying, six or seven radiolabelled pesticides were applied near the base of a 20 x 20 cm plate and developed ten cm with water by ascending chromatography. Pesticide movement was visualized by autoradiography. Movement was expressed by the conventional R designation, although this referred to the front of pesticide movement rather than its maximum concentration. The soil TLC data are most appropriately compared with other mobility data which indicate the depth to which an organic chemical may be

leached. The ranking of pesticides in order of mobility is in good agreement with general trends previously reported.

Absolute movement on soil TLC plates cannot be transposed directly to field or soil column experiments. Since soil structure in the TLC system is considerably more homogeneous that in most other systems, band spreading will be somewhat less than in field or column regimes. Flow rates are also higher than those occurring naturally. For example, infiltration into Hagerstown silty clay loam was equivalent to rainfall of about 1.2 cm/hr (Helling 1970). High flow rates are usually associated with increased mobility, as later correlations (Helling 1968) bore out. In spite of these problems, monitoring data utilizing certain reference chemicals has provided the necessary information to relate soil TLC data to column and field data. In general, Helling and Turner (1968) indicated that soil TLC offered a rapid, simple, and inexpensive procedure for establishing a general mobility classification of pesticides and organic chemicals.

Simple chromatographic theory can be used to correlate adsorption coefficients with soil TLC R_f values. If chromatographic movement through a soil column is treated according to the distillation theoretical plate theory (Block et al. 1958, Martin and Synge 1941), a formula for R_f is obtained in terms of the relative cross-sectional areas of the liquid and solid phases and partition of a chemical between solid and liquid phases (Hamaker 1975):

$$R_f = A_L/(A_L + A_s) = 1/[(1+ \alpha\ (A_L + A_s)] \qquad (2)$$

where A_S and A_L are cross-sectional areas of solid and liquid phases and α is the ratio of volume concentration in the solid phase to that in the liquid phase. For saturated conditions which will be assumed for a soil plate, $A_L + A_S = A$ (cross-sectional area), this can be written:

$$R_f = 1/[(1 + \alpha\ (A/[A_L - 1])] \qquad (3)$$

When reexpressed in terms of the pore fraction of the soil θ, density of soil solids (d_s), and a soil adsorption coefficient K, this equation becomes:

$$R_f = [(1 + K(d_s)(1/\theta^{2/3}-1)]^{-1} \qquad (4)$$

This ratio, A/A_L, is set equal to $1/\theta^{2/3}$ by analogy to the treatment of soil diffusion by Millington and Quirk (1961) where it serves to correct for the tortuosity of flow through the porous medium. In this case, it serves to relate the pore volume to the cross sectional area of the liquid phase in a saturated soil. In general, equation 4 has shown that an inverse relationship exists between the soil adsorption coefficient K and R_f (Hamaker 1975).

Riley (1976) presented a general relationship between the soil/solution distribution coefficient K and the depth of pesticide leaching. Relating the data of Riley (1976) with the R_f values of Helling (1968, 1971a, 1971b, 1971c) and the average K values of Goring and Hamaker (1972) for selected pesticides, the general relationship shown in Table 1 was developed between the soil/solution partition coefficient, R_f, and soil mobility.

4.2 Rationale for Selection of Soil TLC

A number of laboratory tests, the soil thin layer chromatography, soil adsorption isotherm, and soil columns have been developed to obtain an estimate of a chemical's leaching potential (Hamaker 1975). Soil TLC is the first of several tests with will be recommended for use. It is the least expensive of the available tests which measures leaching potential,is widely used, and is the test most amenable to standardization at the present time.

The soil TLC offers many desirable features. First, mobility results are reproducible. Mass transfer and diffusion components are distinguishable. The method has relatively modest requirements for chemicals, soils, laboratory space, and equipment. It yields data that are amenable to statistical analyses. A chemical extraction-mass balance procedure to elicit information on degradation and chemical transformations occurring at colloid interfaces can be incorporated into this test. The ease with which the R_f and mass balance are performed will depend

Table 1. The General Relationship Between the Soil/Solution Partition Coefficient K, R_f and Soil Mobility

K	R_f	Mobility class	Distance surface applied chemical may leach
0.1.....	0.95	Very Mobile.....	
1.....	0.60	.....	Much of chemical leaches through top 20 cm soil into subsoil
10.....	0.25	Mobile.....	Much of chemical leached Into soil but peak concentration in top 20 cm soil.
10^2.....	0.10	Low mobility.....	Only small amount of leaching and peak concentration normally In top 5 cm soil
$10^{2.5}$.....	0.00	.....	
10^3.....	0.00	.....	
10^4.....	0.00	Immobile.....	No significant leaching.

upon the physical/chemical properties of the test chemical and the availability of suitable analytical techniques for measuring the chemical.

4.3. Rationale for Selection of Experimental Conditions and Procedures

The papers by Helling (1968, 1971a, 1971b, 1971c) and Helling and Turner (1968) were the basis of this test standard. The soil and colloid chemistry literature and the analytical chemistry literature substantiates the experimental conditions specified in the suggested standard as accepted, standard procedures. A few of these conditions will be discussed in greater detail below.

Soil TLC can be used to determine the soil mobility of sparingly water soluble to infinitely soluble chemicals. In general, a chemical having a water solubility of less than 0.5 ppm need not be tested since the literature indicates that these chemicals are, in general, immobile (Goring and Hamaker, 1972). However, this does not preclude future soil adsorption/transformation testing of these chemicals if more refined data are needed for the assessment process.

Soil TLC may be used to test the mobility of volatile chemicals by placing a clean plate over the spotted soil TLC plate and then placing both plates in a closed chromatographic chamber.

Soil TLC was originally designed for use with soils. The literature shows no published use of this method with sediments as the adsorbent phase, probably due to the fact that sediment surface properties change significantly during air drying. The TLC plate with the adsorbent must be air dried before leaching studies can be undertaken.

The Test Rule will specify the minimum number of soils to be tested. This number will depend upon the number of sites of manufacture, distribution, use, and disposal of the test chemical. Since there are currently 10 recognized soil orders in the United States containing soils with wide variations in the amounts and types of clay, organic matter, pH, primary and secondary minerals, structure, texture, and exchange capacity the inclusion of a requirement to test soils representing each major order for each chemical would make this standard prohibitively expensive.

Distilled-deionized H_2O is required in order to minimize competition effects for soil exchange sites by cationic and anionic species normally present in tap and distilled H_2O.

The purity of the test chemical will be specified in the test rule. In general, however, the test chemical should be of the purest grade available. Impurities may produce migration patterns on the TLC plate independent of the parent chemical and may be misinterpreted as transformation products. Transformation product identification is an expensive analytical procedure that

may be unnecessarily required as a result of the presence of impurities.

The sieving of soils will remove the coarse (500-2,000 µm) and medium (250-500 µm) sand fractions. Published testing results showed that removal of a portion of sand had no affect on the mobility of a test compound but aided in achieving a more cohesive uniform soil layer and more reproducible results (Helling 1971a).

Gentle crushing and grinding must be used to reduce soil aggregate size. Fine particles (silt and clay) in excess of the amount originally present may be created if excessive pressure is exerted on the aggregates.

Application of the soil slurry to clean glass plates must be done quickly to prevent particle size segregration. A specific method of soil slurry application was not identified since a number of methods which produce the acceptable layer thickness are in use today.

Replication of the basic experimental unit was necessary in order to estimate the standard deviation of the treatment mean. Three replicates are considered to be the minimum number of replicates for a statistically acceptable estimation. The soils literature indicates that, in general, the standard deviation should be less than 0.01 R_f units for soil TLC.

Since the available literature indicated that pesticide mobility on soil TLC plates did not significatly change when

temperature varied from 2° to 25°C (Helling 1971a), only a room temperature range was suggested.

The Soil Order, Series, and general clay fraction mineralogy data may be found in Soil Survey Reports published after approximately 1970. Pre-1970 reports may not contain mineralogy data. Soil Survey Reports have been issued for most U.S. counties and may be obtained from County Extension Offices; the State Office of the U.S. Department of Agriculture Soil Conservation Service; or the USDA-Soil Conservation Service, Publications and Information Division, P.O. Box 2890, Washington, DC 20013. If mineralogy data are not printed in a report, the State Office of the U.S. Department of Agriculture Soil Conservation Service may be contacted for assistance in obtaining general clay mineral data of a particular soil. The test standard does not require test soil mineral analysis since general clay mineralogy data may already exist for the test soil.

4.4. Replies to Federal Register of March 16, 1979

The Federal Register of March 16, 1979 (EPA 1979) presented a discussion of policy issues, alternative approaches, and test methods under consideration for premanufacture testing. Included in the test methods was a soil thin layer chromatography test method (pgs. 16257-16264). Several comments received on this method will be discussed in this section.

One commentor suggested that it is not possible to determine the adsorption coefficients of organic compounds, having

solubilities less than 0.05 ppm, with any degree of accuracy. Therefore, a 0.5 ppm solubility limit has been adopted in the soil TLC test standard.

Several commentors stated that the 95% extraction efficiency in the mass balance section of the March 16, 1979, standard is too stringent and too costly to implement. An 80 percent extraction efficiency has been adopted in response to this comment and is believed to be a more reasonable indicator of a chemicals propensity to transform.

One commentor suggested that the high pressure liquid chromatography technique HPLC be adopted instead of the soil TLC for soil mobility testing. Sections 2.3., 2.4., and 2.5. of this support document indicate that hydrophobic bonding is an important effect governing soil mobility, but is not the only effect that must be considered for all chemicals. HPLC measures only the hydrophobic bonding potential of a neutral organic chemical. Also section 2.6. indicates that surface transformations may occur but no scheme to predict the occurence of these reactions presently exists. HPLC does not measure this effect. For these reasons, the suggestion was not adopted.

Several commentors stated that the use of soil as the adsorption medium is impractical. A substitute adsorbent such as carbon, clay, humic substances, oxides, and silica and/or alumina plates could be used with several solvents of different polarity. This suggestion was not adopted for several reasons. First, water is the liquid environmental transport media of

concern. Use of different solvents would give little insight into the mobility of chemicals in soils. Second, section 2.4., 2.5., and 2.6. of this document indicate that soil is a heterogeneous mixture of numerous inorganic and organic phases. The published literature does not substantiate the use of one substitute adsorbent as an adequate replacement for soils during mobility tests. Third, the literature does not indicate how data derived from these adsorbents can be extrapolated to soils.

* U. S. GOVERNMENT PRINTING OFFICE : 1980 341-085/3932

5. TSCA/FIFRA/OECD COMPARABILITY

Leaching data are required to support the FIFRA registration of all formulated products intended for terrestrial noncrop, tree fruit/crop, and field/vegetable crop uses, and for terrestrial/aquatic (forest) uses. One preferred technique to support registration is soil TLC. Data derived for the Office of Pesticide Programs will be accepted under TSCA provided chemical mass balance data is submitted and soil physical/chemical property guidance as discussed in the test rule have been met.

The Organization for Economic Cooperation and Development recognizes the importance of determining the mobility of a chemical in soils to assess the potential risk to man and the environment due to the manufacture, distribution, use, and disposal of chemical products. The soil adsorption isotherm was identified by OECD as a potential method for determining the leaching potential of chemicals in soils. This test will be proposed as a standard method in the future.

6. REFERENCES

Adams Jr. RS. 1973. Factors influencing soil adsorption and bioactivity of pesticides. Residue Rev. 47:1-54.

Bailey GW, White JL. 1970. Factors influencing the adsorption, desorption and movement of pesticides in soil. Residue Rev. 32:29-92.

Block RJ, Durrum EL, Zweig G. 1958. A manual of paper chromatography and paper electrophoresis. Second Edition. Academic Press, N.Y.

Buch RR, Ingebrigtson DN. 1979. Rearrangement of poly-(dimethyl/siloxane) fluids on soil. Environ. Sci and Technology 13:676-679.

Buckman HO, Brady NC. 1969. The nature and properties of soils. London: The Macmillan Company

Enviro Control, Inc. 1980. Cost analysis methodology and protocol estimates: environmental standards. Submitted to the Office of Regulatory Analysis, U.S. Environmental Protection Agency, Washington, DC

Goring CAI, Hamaker JW eds. 1972. Organic chemicals in the soil environment. Vol. I & II. New York: Marcel Dekker, Inc.

Guenzi WD ed. 1974. Pesticides in soil and water. Madison, WI: Soil Science Society of America, Inc.

Hamaker JW 1975. The interpretation of soil leaching experiments. In Haque R and Freed VH eds. Environmental science research Vol. 6: Environmental dynamics of pesticides.

Helling CS 1968. Pesticide mobility investigations using soil thin-layer chromatography. Amer. Soc. Agron. Abstracts p. 89.

Helling CS, Turner BC. 1968. Pesticide mobility: Determination by soil thin layer chromatography. Science 162:562.

Helling CS. 1970. Movement of s-triazine herbicides in soils. Residue Review 32:175-210.

Helling CS. 1971a. Pesticide mobility in soils I. Parameters of soil thin layer chromatography. Soil Sci. Soc. Amer. Proc. 35:732-737.

Helling CS. 1971b. Pesticide mobility in soils II. Applications of soil thin layer chromatography. Soil Sci. Soc. Amer. Proc. 35:737-743.

Helling CS. 1971c. Pesticide mobility in soils III. Influence of soil properties. Soil. Sci. Soc. Amer. Proc. 35:743-748.

Martin AJP , Synge RLM. 1941. A new form of chromatogram employing two liquid phases. Biochem. J. 35:1358.

Millington RJ and Quirk JP 1961. Permeability of porous solids. Trans. Faraday Soc. 57:1200.

Mingelgrin U, Saltzman S, Yaron B. 1977. A possible model for the surface induced hydrolysis of organophosphorus pesticides on kaolinite clays. Soil Sci. Soc. Amer. Jour. 41:519-523.

Mortland MM, Halloran LJ. 1976. Polymerization of aromatic molecules on smectite. Soil Sci. Soc. Amer. Jour. 40:367-370.

Riley D. 1976. Physical loss and redistribution of pesticides in the liquid phase. In: British Crop Protection Council Symposium Proceedings. p. 109-115.

Shearer RC, Letey J, Farmer WJ, Klute A. 1973. Lindane diffusion in soil. Soil Sci. Soc. Amer. Proc. 37:189-193.

Stevenson FJ. 1973. Organic matter reactions involving pesticides in soil. In Bound and conjugated pesticide residues. ACS Symposium Series Monograph 29/1976.

Wang TDC, Li,SW. 1977. Clay minerals as heterogeneous catalysts in preparation of model humic substances. Z. Pflanzenernaehr. Bodenkd. 140:669-676.

Wang TSC, Li SW, Ferng YL. 1978. Catalytic polymerization of phenolic compounds by clay minerals. Soil Sci. 126:15-21.

White JL. 1976. Determination of susceptibility of s-triazine herbicides to protonation and hydrolysis by mineral surfaces. Arch. Environ. Contam. Toxicol. 3:461-469.

Yaron B. 1978. Some aspects of surface interactions of clays with organophosphorus pesticides. Soil. Sci. 125:210-216.

REPORT DOCUMENTATION PAGE	1. REPORT NO. EPA 560/11-80-027	2.	3. Recipient's Accession No.
4. Title and Subtitle: Support Document, Test Data Development Standards, Physical/Chemical & Persistence Characteristics: Density/Relative Density, Melting Temperature, Vapor Pressure, Octanol/Water Partition Coefficient, Soil Thin Layer Chromatography.			5. Report Date: October 1980 6.
7. Author(s) Geo. T. Armstrong, Robert H. Brink, Asa Leifer, James Dragun.			8. Performing Organization Rept. No.
9. Performing Organization Name and Address: U. S. Environmental Protection Agency, Office of Pesticides & Toxic Substances, Office of Toxic Substances, Exposure Evaluation Division, 401 M Street, S.W., Washington, D.C., 20460			10. Project/Task/Work Unit No. 11. Contract(C) or Grant(G) No. (C) (G)
12. Sponsoring Organization Name and Address: U. S. Environmental Protection Agency, 401 M Street, S.W., Washington, D.C., 20460			13. Type of Report & Period Covered 14.

15. Supplementary Notes

This is a Support Document to a Proposed Rule for Environmental Test Standards under the Toxic Substances Control Act (TSCA), published in the Federal Register October 1980

16. Abstract (Limit: 200 words)

This technical Support Document provides the rationale for the development of test standards to develop data on density/relative density, melting temperature, vapor pressure, octanol/water partition coefficient, and soil thin layer chromatography of chemical substances. EPA will use the data on these physical/chemical characteristics to evaluate the manner and extent of environmental transport, fate and places of deposit as an aid in assessing health and environmental effects of chemicals under TSCA. For density/relative testing, an analysis is given of available methods of determining this property of particular classes of materials with different physical characteristics. For melting temperature testing, available methods are analyzed in terms of materials with different physical characteristics. For vapor pressure, two procedures are given, the isoteniscope procedure for pressures of 0.1 to 100 kPa and a gas saturation (transpiration) procedure for pressures of 10^{-5} to 10^{3} Pa. The Knudsen effusion procedures are also given. How to determine the numerical values of the octanol/water partition coefficient are given. Soil thin layer chromatography, an experimental method for determining the relative mobility of organic chemicals in soils, is discussed, including scientific aspects of soil leaching, economic aspects of the method, and scientific history and the rationale for selection of experimental conditions for this method.

17. Document Analysis a. Descriptors

Environmental Transport
Environmental Exposure Pathway
Chemical Tests
Chemical Analysis
Octanol/Water Partition Coefficient
Adsorption
Leaching
Soil Chemistry

b. Identifiers/Open-Ended Terms

Density, Specific Gravity; Melting Point, Temperature; Gas Saturation Procedure, Isoteniscope Procedures, Gas Transpiration Procedures; Bioaccumulation, Ecological Concentration, Partitioning Measurement; Soil Mobility, Soil Thin Layer Chromatography, Soil Tests.

c. COSATI Field/Group 07D Chemistry, Physical Properties

18. Availability Statement	19. Security Class (This Report)	21. No. of Pages
This document is available for general public release.	none	ca. 200 pp
	20. Security Class (This Page) none	22. Price

(See ANSI-Z39.18) See Instructions on Reverse OPTIONAL FORM 272 (4-77) (Formerly NTIS-35) Department of Commerce

best copy available

United States
Environmental Protection
Agency

Official Business
Penalty for Private Use
$300

Special Fourth-Class Rate
Book
Postage and Fees Paid
EPA
Permit No. G-35

Washington DC 20460